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BULLETIN

OF THE NEW YORK ACADEMY OF MEDICINE

Original Articles

by

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CLINICAL RESEARCH MEETING

SECOND SERIES JULY 1949 VOL. 25, NO. 7

BULLETIN OF THE NEW YORK ACADEMY OF MEDICINE

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MAHLON ASHFORD, *Editor*

Published Monthly by THE NEW YORK ACADEMY OF MEDICINE
2 East 103 Street, New York 29, N. Y.

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BULLETIN OF
THE NEW YORK ACADEMY
OF MEDICINE

JULY 1949

IRON METABOLISM AND HEMOCHROMATOSIS*

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THIS review will be divided into four parts. In the first part we will enumerate and classify the iron compounds of the body. In the second part we will describe the properties of some of these iron compounds. In the third part, the properties and the hypotheses concerned with the regulation of iron absorption by the gastro-intestinal mucosa will be discussed, and in the last part we will consider certain of the aspects of iron metabolism as they might be related to hemochromatosis.

I. IRON COMPOUNDS OF THE BODY

No detailed analyses exist for all the different iron compounds of the human. However, by incorporating various data from the literature, an approximate notion may be obtained of the distribution of total body iron among these various compounds. The iron compounds of the body may be divided into two groups. The first group will include the iron porphyrin or heme compounds and in the second group will be included the non-heme iron compounds (Table I).

The iron porphyrin heme compounds will be considered here very

* From the Laboratories of The Rockefeller Institute for Medical Research, New York. Read December 3, 1948 in the Friday Afternoon Lecture Series of The New York Academy of Medicine.

TABLE 1: APPROXIMATE COMPOSITION OF THE IRON-CONTAINING COMPOUNDS IN THE HUMAN (70 KILO MAN)*

Compounds	*g.*	*Fe in g.*	*Per cent of total Fe*	*Reference No.*
Iron porphyrin (heme) compounds:				
Blood hemoglobin	900	3.0	60-70	(2)
Muscle hemoglobin (myoglobin)	40	0.13	3.0-5.0	(1, 2)
Heme enzymes:				
Cytochrome c	0.78	0.004	0.1	(2)
Cytochrome a_3, b	—	—	—	
Catalase	—	—	—	
Peroxidase	—	—	—	
Non-heme iron compounds:				
Iron monomolecularly dispersed:				
Ferrous ion	—	—	—	
Siderophilin (Fe+++)	10.0	0.004	0.1	(4, 5, 6, 10)
Iron in micellar ferric hydroxide units:				
Ferritin	2.0-4.0	0.4-0.8	15	(7, 8)
Non-crystallizable ferritin	—	—	—	(7, 8)
Hemosiderin	—	—	—	(7, 8)
Total Iron		4—5	100	(1)

briefly. The essential function of the heme compounds when they are associated with particular proteins is to make oxygen available to the cell. We may best see how this is accomplished by following oxygen on its path into the cells. The oxygen in the lungs is taken up by the hemoglobin of the erythrocytes and transported to the tissues. In red striated muscle and heart muscle the oxygen is temporarily stored in muscle hemoglobin or myoglobin. Then the oxygen passes on in the cell to cytochrome oxidase (cyt.a_3), a heme enzyme which activates the oxygen to accept electrons. Other heme enzymes, as cytochromes c and b, serve to transport or shuttle electrons from organic compounds to the activated oxygen on the cytochrome oxidase. Another kind of heme enzyme, peroxidase, can activate H_2O_2 to act as a strong oxidant, and still another heme enzyme, catalase, can act to destroy toxic accu-

* The striated muscles of various animals, although not other tissues, appear to contain two non-heme iron fractions according to Dreyfus, Schapira and Leau, *Bull. Soc. Chim. Biol., 30,* 99 (1948); one fraction, extractable by pyrophosphate may represent 30-90% of the total non-heme iron depending on the species. Assuming non-heme iron levels for muscle to be the same in human as in beef then for a 70 kilo man the total skeletal muscle would contain about 50 mg. and the heart about 12 mg. non-heme iron.

mulations of H_2O_2 which may be generated in the cell during these processes of oxidation with O_2. It should be noted that the amount of iron in all of the heme enzymes of the cells is extremely small, being only a few tenths of one per cent of the total iron of the body. This is in marked contrast to the amount of iron in the hemoglobins of the red cell and muscle which together account for 65-75 per cent of the total body iron, assuming an average of 4.5 g. for total body iron.[1-8]

The second group of iron compounds, namely, the non-heme group, is the one with which we shall be primarily concerned this afternoon. This group includes inorganic iron compounds and certain iron protein compounds (Table I). These compounds may be subdivided into two classes. The first includes the compounds of iron where the iron atoms are not in clusters—i.e. where the iron atoms are monomolecularly dispersed; the two substances that may be placed in this category are ferrous iron itself as a free ion and siderophilin, the β_1 serum globulin,—that is, the transport form of iron in the blood stream.

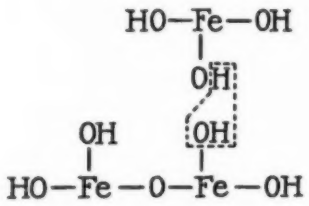
The second class of non-heme compounds contains iron in the form of ferric hydroxide units or clusters of a special character. The ferric hydroxide units are contained in ferritin, non-crystallizable ferritin and hemosiderin. These compounds containing ferric hydroxide units, serve as storage forms for iron. As one might expect from storage compounds, their content varies greatly from individual to individual.

Having briefly surveyed the various iron compounds of the body, let us now summarize the table on the approximate content of these compounds. Let us assume a total of some 4-5 grams for the iron in the body of a 70 kilo man. Then the total hemoglobin iron, i.e. in hemoglobin and myoglobin, accounts for the greatest fraction, 65-75 per cent, and ferritin iron makes up the next greatest fraction, about 15 per cent. Together these constitute about 85-90 per cent of the total iron. The other known constituents make up less than one per cent of the iron. There still remain, to be accounted for, about 10-15 per cent of the total iron of the body. Whether this latter amount of iron reflects errors in estimations, especially of total body iron or whether other compounds of significant iron content remain yet to be discovered, it is not possible to say.

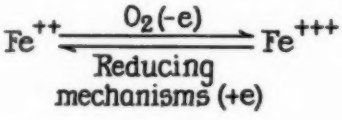
II. Properties of the Non-Heme Iron Compounds

Before discussing the properties of the non-heme iron compounds

two important properties of inorganic iron may be noted. The first is that iron may exist in the body in two states of oxidation, that is, either in the ferrous or the ferric state. The second is that the solubility of ferric ions is extremely slight. In contrast to the solubility of ferric ions, the solubility of ferrous ions, although low at body pH, is of significant magnitude. The reason for the very slight solubility of ferric iron is that the ferric iron is converted to ferric hydroxide at the pH of the body, and several ferric hydroxide molecules have a very great tendency to come together and split out water among them. In other words, the ferric hydroxide molecules aggregate or polymerize in three dimensions very readily into clusters or micelles of ferric hydroxide, thus:



Ferrous iron. We may look upon ferrous ions as the starting point from which all iron compounds of the body originate. Ferrous ions are only slightly soluble at body pH, but sufficiently soluble so that they may be reasonably considered to be an important metabolic intermediate. The solubility of ferrous hydroxide is equivalent to a concentration of 25 γ Fe per cent, but the ferrous iron may be made more soluble by the presence of bicarbonate or by loose complex formations with amino acids, etc. Unfortunately there is no adequate method for measuring the inorganic ferrous iron in the tissues. The reason for this difficulty becomes apparent when we realize that the level of ferrous iron is due to a dynamic equilibrium. In the presence of oxygen, ferrous iron tends to become oxidized very readily to the ferric form. There is, however, a tendency to reverse this process. The ferric iron is converted back to the ferrous form by the reducing mechanisms of the cell:



So the level of ferrous iron depends on the rate at which it becomes autoxidized directly by oxygen (or indirectly by oxidation catalysts) and the rate at which it is reduced back to the ferrous form. An understanding of the factors that are involved in the maintenance of this steady state, we believe, is basic to an understanding of hemochromatosis.

The second compound of this group of monomolecularly dispersed iron is siderophilin which serves as a transport form for iron (Cohn fraction IV-7). This protein molecule has been crystallized. It is a pseudo-globulin of molecular weight 90,000 that can carry two iron atoms in the ferric state. The protein is itself colorless. Schade and co-workers[4, 10] have found that when ferrous iron is added to the colorless protein the iron becomes autoxidized to the ferric state and in the presence of CO_2 attaches to the protein. The protein now takes on a salmon-pink color. This pink complex contains one CO_2 for each ferric atom.[10] The iron can be removed from the protein by adding a reducing substance to convert it to the ferrous state; or the ferric iron can be removed by lowering the pH to 4.6 (i.e. a less physiological expedient). It is important to note that no enzyme need be directly involved in the addition or removal of the iron to or from this protein. In the blood stream this protein is normally only about one-third saturated with iron, the serum iron level being about 100 γ per cent; when saturated with iron the serum iron level is about 300 γ per cent. It has been shown that the iron normally carried in the serum is carried exclusively by this protein. This protein may also carry copper, presumably at the same spots on the protein that would carry the iron.[6] When the protein has copper on it, if one adds an iron salt, the iron will displace the copper. The constancy of the serum iron level is also indicative of a steady-state process. It therefore reflects various aspects of iron metabolism. We shall discuss this feature later.

The second class—the colloidal or micellar iron compounds—contain aggregates or polymers of ferric hydroxide. One of these substances is ferritin, and another of these is hemosiderin.[7, 8]

Ferritin is a remarkable compound, first discovered by Laufberger in 1937. It is most readily isolated from horse spleen which is one of the richest sources of this substance. Ferritin is a brown iron-containing protein which can be crystallized readily by means of cadmium sulfate. In the crystallized state it may contain as much as 23 per cent dry weight of iron. One may remove the iron without damaging the

protein and the colorless protein, now called apoferritin, also will crystallize when $CdSO_4$ is added. So the property of crystallization is a property of the protein. The protein apoferritin has a molecular weight of 460,000.

Figure 1 is a hypothetical reconstruction of what a crystal of ferritin, containing 23 per cent iron, might look like. Here we see the somewhat ellipsoid apoferritin protein molecules and in the spaces we see ferric hydroxide micelles or clusters (cross-hatched spheres in the illustration) attached to the surface of the protein. Some cadmium atoms are represented as binding together the protein molecules possibly by coördinating certain amino acid groups on the surface of the two protein molecules.

The ferric hydroxide micelles of ferritin are not uniform in size, but are in general small enough to permit crystallization. Their average composition indicates that they contain about 1 PO_4 for every 9 iron atoms, i.e. $[(FeOOH)_8.(FeOPO_3H_2)]$. Theoretically, three types of ferric compounds are possible on the basis of the number of unpaired electrons which ferric iron may possess in the outermost electron shell. The ferric compounds may have either one, three, or five unpaired electrons per iron atom. The most common type of ferric compounds are those containing one and five unpaired electrons. An example of the type having five unpaired electrons is ferric chloride, or the iron in the transport compound, siderophilin; of the type having one unpaired electron an example is the iron in ferricyanide or in methemoglobin cyanide. These types are distinguished experimentally by making magnetic measurements to determine the number of unpaired electrons per iron atom. When ferritin was first measured by Michaelis, using the magnetic method, it was found that ferritin contained three unpaired electrons. The type having three unpaired electrons per iron atom is very rare. In the organism, ferritin iron can be distinguished from all other iron compounds since only ferritin iron has three unpaired electrons. Now the few experiments that have been done on the formation of these micelles or polymers indicate that these iron hydroxide units are polymerized through the mediation of some enzyme activity. It seems likely also that the individual micelles are attached to the apoferritin molecule through enzyme mediation. Support for the idea that an enzyme is involved in the formation of these micelles is the peculiar three-unpaired electron type, a type which we have been

unable to duplicate by *in vitro* experiments.

Ferritin may be considered to have three functions. Its primary function is that of a normal iron storage protein of the body. Many different tissues of the body appear to be able to make ferritin. Normally, ferritin may be isolated from the spleen, liver and bone marrow of a number of animals and in lesser amounts from testicles, ovary, kidney and pancreas. Agner has reported traces of ferritin in horse erythrocytes.[11] When the iron arriving in a tissue becomes relatively great, even a tissue like heart muscle which normally may contain very little or no ferritin, will be found to contain ferritin. Ferritin produced by the cells of one tissue, say the liver, is immunologically identical with ferritin produced by cells of other tissues of the body. From the average human liver one may isolate about ½ to 1 gr. dry weight of ferritin; because of the inefficiency of isolation it may be estimated that twice this amount may be present.*

The second function of ferritin is related to the regulation of iron absorption. The mucosal cells of the gastro-intestinal tract control the amount of iron absorbed. The formation of ferritin in these mucosal cells is in some way connected with the production of a mucosal bloc in the mucosa, preventing excessive absorption of iron. Vosburgh and Flexner[12] have recently found ferritin in the human placenta and it seems likely that the placenta may also contain a mechanism similar to that of the intestinal mucosa, for the regulation of the iron absorbed by the embryo from the mother.

A third action of ferritin, and one which may be important in an understanding of irreversible shock, hypertension, edema, etc., has been recently revealed by Mazur and Schorr.[13] This is concerned with the vasoconstrictor action of adrenalin. By the method of Zweifach and Chambers[14] one may observe under the microscope the effect of the vasoconstrictor action of adrenalin on the terminal arterioles of the rat meso-appendix. When extremely small amounts of ferritin are added in the presence of adrenalin, the vasoconstrictor action of adrenalin is inhibited. In normal serum, ferritin appears to be absent or at least below the threshold sensitivity of this delicate test. However, in traumatic shock or in patients with liver cirrhosis, traces of ferritin may escape from the injured cells into the serum. The presence of the

* One of the highest and one of the lowest values for ferritin were found in two cases of leukemia. In one case of cirrhotic liver the ferritin crystals which were obtained in small quantity were pale yellow in color and contained only 5 per cent instead of the usual 20-23 per cent iron.[9]

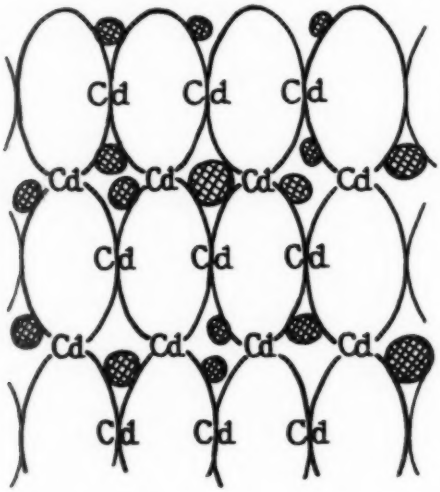


Fig. 1—Hypothetical reconstruction of a cross section of a ferritin crystal

ferritin in such serum can be demonstrated by its vasodepressor activity in the meso-appendix test. That it is ferritin and not another component of the serum which is active can be shown by adding antibody directed against ferritin. Then no vasodepressor activity is observed, that is, the vasodepressor activity of ferritin is counteracted by neutralization with its specific antibody. The effect of the ferritin is not due to its iron content since apoferritin will produce the same vasodepressor action as ferritin. The rat will respond to ferritin obtained from other species than its own, so it seems that there are common groupings on the protein that are responsible for the vasodepressor activity. The practical consequences of such a vasodepressor activity of ferritin and its control are clearly apparent.

When ferritin is crystallized out, there still remains in the mother liquor a brown pigment, called non-crystallizable ferritin. This has the characteristics of ferritin except that it will not crystallize. The evidence suggests that in this fraction the iron hydroxide micelles on the surface of the protein may be too large to fit into the crystal lattice (Fig. 1).

Hemosiderin is a granular substance which contains clusters of iron hydroxide units mixed with some protein material. These granules are

large enough to be microscopically visible and they stain for iron. The hemosiderin granules, separated from horse spleen, may contain as much as 35 per cent by weight of iron. This iron seems to have the same magnetic properties as ferritin iron or to deviate only slightly from it, indicating that the way the iron hydroxide units polymerize is probably identical with the way ferritin iron is polymerized. One might picture the formation of hemosiderin as an abnormal stage of iron deposition beyond that of ferritin. Ferric hydroxide micelles attached to ferritin, instead of consisting of small polymers might grow abnormally large. At the same time, cross-links between these enlarged ferritin molecules could take place (i.e. a cross-link could be formed by splitting out H_2O between two ferric hydroxide polymers on the surface of two separate ferritin molecules). It should be noted that the iron contained in these large clusters is less readily available to the body than is the iron of ferritin. The iron of ferritin is in minute clusters with relatively large surface area, such iron being more readily brought into solution by the reducing processes of the cells.

A reasonable view regarding hemosiderin formation is that if iron enters too rapidly into a tissue for apoferritin synthesis to keep pace with the entry of iron, or if a tissue is already saturated with ferritin, then this iron gets heaped up into granules. It is generally true that when one finds hemosiderin granules, one will find a high concentration of ferritin. However, one should not consider the hemosiderin granule as a normal storage form merely because it is microscopically visible. Ferritin is not microscopically visible nor can it be readily detected by iron stains because the ferritin is too diffuse in the cells, yet the iron of ferritin may be present in a decidedly higher concentration in the cell than is represented by the visibly stainable iron of the hemosiderin granules.

There is another form of microscopically stainable iron that might be confused with hemosiderin. This is the iron released from denatured hemoglobin. When red cells are damaged and engulfed by phagocytes, some of the material of the red cells is temporarily converted to denatured hemoglobin granules. These granules, lying in the digestive vacuoles of the phagocytes, first appear yellow-brown, the color being due to ferric hydroxide which is stainable, and to yellow bile pigment. Within 3-4 days the iron staining may disappear although some of the partially digested protein granules may still be tinged pale yellow due

to absorbed bilirubin. The iron may be carried away from the phagocytes by way of the blood stream and stored for example, in the liver. Or cells adjacent to these phagocytes may absorb the iron and convert it to ferritin iron if the cells have such an ability. For example, as seen in the dog, the slight hemorrhage resulting from release of a graafian follicle gives rise to a brown area in the vicinity of the corpus luteum from which ferritin has been crystallized.

In one experiment, an abnormal form of iron has been encountered. Normally, as shown in dogs,[15] when a ferric salt is injected into the blood stream, it is absorbed rapidly by the liver and its five unpaired electron type is converted to the three unpaired electron type characteristic of the ferric hydroxide micelles of ferritin. In one instance this conversion did not take place and it can only be concluded that the enzymes which normally would convert this ferric iron were not functioning properly, that is, the dog may have been ill. Ferric iron, which is not handled properly, is toxic, and we shall discuss further on how a similar situation, arising in hemochromatosis might result in liver damage.

III. Regulation of Iron Absorption

In contrast to the traces of iron that are needed to satisfy the heme enzyme requirements of simple forms like yeast, one may place the relatively enormous iron requirement of the mammal. This high iron requirement is due to the high content of hemoglobin in the mammal. In the body the iron contained in the hemoglobin of the red cells is about 3.0 grams. This is about one thousand times more iron than is used for the manufacture of all the heme catalysts of the body combined. In order to obtain an adequate amount of iron for hemoglobin synthesis, mechanisms had to be developed by the vertebrates for accumulating and regulating these biologically huge quantities of iron.

Absorption in the form of ferrous iron. The stomach has an important action on the iron ingested with the food. The stomach is acid, and this acidity or lowered pH has two effects. First, it converts colloidal ferric hydroxide of foodstuffs into monomolecularly dispersed ferric ions. Second, at acid pH certain substances in the food act to reduce the ferric ions to the ferrous form. For example, ascorbic acid or cysteine or the -SH groups of proteins in food will not reduce ferric salts at pH 7, but reduction can already be observed at pH 5.

The reduction of iron to the ferrous form is of importance because it has been clearly demonstrated that it is in the ferrous form that the iron is absorbed.[16]

Although iron may be absorbed all along the gastro-intestinal tract, very little is absorbed normally except in the duodenal region.[1,17] In the acid stomach the ferrous iron which is formed by reduction, is stable, that is, it is not readily autoxidizable by oxygen. In the duodenum there is present a most active region of absorptive surface and a pH which is still not much above 5. The region just below the pyloric sphincter will therefore be most effective in absorption of ferrous iron. As the iron moves further down the intestinal tract the digestive juices of the pancreas, bile and the intestinal mucosa tend to raise the pH to 6-7. At this higher pH the ferrous iron becomes rapidly autoxidizable and will be converted to ferric hydroxide. Normally with the relatively low content of iron coming in with the food, say 2 to 10 mg. Fe per day, the main absorptive region may be limited to a very short segment below the pyloric sphincter. If a larger dose of iron is given, say 50-100 mg. Fe, a larger region of the duodenum may be involved in absorption, and with still larger doses of iron as is suggested for use in iron deficiency anemia (300 mg. Fe^{++}, 3-4 times daily), more of the intestinal tract, perhaps even the large intestine and stomach, may absorb significant amounts of iron. Even under conditions of anemia and this high dosage, only about 15 mg. of a total of 300 mg. Fe^{++} may be absorbed. When a large dose of ferrous iron is fed to a hypochromic anemic the serum iron may rise to saturation level in 6 hours and drop down in 4 hours more to its original low level.[18]

When a dose of iron is first increased, it may be found that the efficiency of absorption of this dose will be relatively high because a new region of mucosa comes to take part in the absorption. However, it should not be considered that this efficiency remains high if this higher dose continues to be fed since a mucosal bloc would be rapidly established in this new region and again absorption would decrease.

Properties of the mucosal cell in the regulation of iron will now be discussed. In general, the organism may regulate a substance taken into the body by way of the gastro-intestinal mucosa in two ways. The organism may either absorb all of the particular substance such as K^+ or Na^+ into the blood stream and excrete the excess; or the organism may develop a mechanism for regulating the amount of the

substance which is to be absorbed. In the case of iron, a regulatory mechanism of great nicety, residing in the mucosa of the gastro-intestinal tract, was developed.* Two features of this regulation may be noted. In the first place, the mucosa behaves as if it knew how to maintain a low, relatively steady rate of absorption under normal conditions. In the second place, when the red cells are below their normal number, i.e., in anemia, the mucosa behaves as if it knew that it must respond by absorbing iron with greater efficiency.[1]

For example, let us consider what is the amount of iron absorbed under normal conditions. An average meal may contain approximately 5 mg. Fe, of which only about 0.5 mg. may be absorbed. In other words, for this dose of iron the efficiency of absorption is about 10 per cent. This would mean that a maximum of about 1.5 mg. Fe per day would be absorbed. It has been estimated that the body may lose a total of about 1 mg. or less of iron per day.[19] This loss is mainly due to the sloughing off of cells from the gastro-intestinal tract, including traces of iron lost in the bile and still smaller quantities lost in the urine. Thus the net gain by the body may be as much as 0.5 mg. of iron per day. The accumulation of 0.5 mg. of iron per day would be sufficient to care for the needs of even a growing child.[1] Considering the very approximate nature of these calculations one may say that on an average diet, the net gain in iron would serve for the manufacture of about 300-600 cc. of blood per year.

Suppose now that blood were lost by hemorrhage and that an anemia ensued. Then one would find, for example, that instead of only 10 per cent of the iron being absorbed, 50 per cent would be absorbed.

Let us next suppose that even though no anemia existed, the efficiency of iron absorption, instead of being 10 per cent, were increased to 30 per cent; then about 3.5 mg. of iron per day would accumulate. The result of this increased efficiency would be that over a 20-year period some 30 gm. of iron would be deposited in the body. Such an amount of iron is sufficient to produce symptoms of the disease called hemochromatosis. We see from this example, that whether the process of absorption is to be considered normal or abnormal may depend on relatively small differences in the level of efficiency with which iron is absorbed over a long period of time.

* Recently, Mitchell and Hamilton, *J. Biol. Chem.*, *178*, 345 (1949) have reported an average daily excretion of 6.5 mg. Fe in male human sweat. This surprising finding, which according to these authors might make unnecessary the assumption of a mechanism for limiting iron absorption, awaits confirmation.

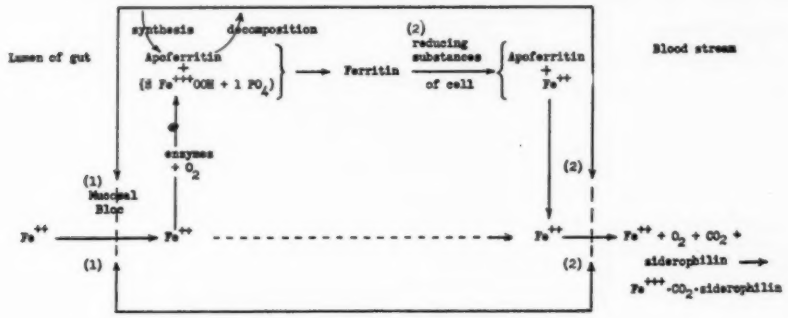


Fig. 2. Hypothesis of the regulation of iron absorption by the mucosal cell

(1) The amount of ferrous iron moving into the cell is regulated by a mucosal bloc related to the presence of ferritin.

(2) The amount of ferrous iron moving into the blood stream depends on the relative redox level of the cell and this in turn is a function of the amount of O_2 supplied by the blood stream.

We shall now consider the facts that reveal a regulatory mechanism for absorption of iron in the gastro-intestinal mucosa. (The diagram of Figure 2 may be useful in the following discussion.)

1. It can be shown that once iron gets into the organism it can no longer get out. For example, Widdowson and McCance[20] demonstrated in 1937 that iron injected into the blood stream in large doses was not excreted by way of the urine or feces. More recent experiments with radioactive iron have confirmed this observation. A dramatic demonstration of this fact is seen in a patient with hemolytic anemia who had some 200 blood transfusions over a period of several years. The iron from all of these broken-down red cells totalled about 25 gm. The patient developed hemochromatosis and died of the effects of this relatively huge deposit of iron in all of the tissues of the body including the heart muscle.

It follows, if iron cannot be excreted after absorption, that the movement of iron from the intestinal tract into the mucosal cell must be always in one direction, i.e., no iron can move out of the mucosal cell into the intestinal tract. So one of the properties that may be deduced from the non-excretion of iron is that the *mucosal cells must provide a one-way transfer of ferrous iron into these cells.*

2. Next it can be shown, that the mucosal cell must contain a mechanism which will regulate the amount of iron finally entering the blood

stream. The experiments of Hahn and others at Whipple's laboratory, performed on dogs with radio iron as tracer, have revealed this fact.[1,21] If radioactive iron is fed to a normal dog, relatively little iron is absorbed. If radio iron is fed to a chronically anemic dog the iron is absorbed in an amount 5-20 times the amount absorbed by the normal dog. Other workers have confirmed this finding in humans. This experiment tells us first, that an *anemia can increase the absorption of iron into the blood stream.* Secondly, it tells us that *in the normal there is a resistance or mucosal bloc to iron absorption.*

3. Another experiment of Hahn and coworkers[1,21] shows that there is a relationship between this resistance, or mucosal bloc, and the storage iron. When a normal dog was bled severely to produce an acute anemia the rate of iron absorption was found to be normal during a period of about a week. Only after this time was iron absorbed at a greater rate, that is, there was a lag in response to the anemia. This experiment suggests that during this early period, immediately after bleeding, the storage iron in the form of ferritin was being depleted to be utilized in the synthesis of new erythrocytes. Not only was the ferritin depleted in the liver, spleen and marrow, but the ferritin in the mucosal cells of the gastro-intestinal tract was also broken down. If the lag represents depletion of ferritin in the mucosal cells, then one should be able to demonstrate a change in ferritin content of the mucosal cells in response to the feeding of iron.

4. That the level of ferritin in the mucosal cells readily responds to the feeding of iron has been demonstrated in the guinea pig. In this animal ferritin is normally present only in traces in the mucosa. When high doses of ferrous iron are fed, ferritin is found to increase, especially in the duodenal region and then after a few days to decrease. In about 3-4 hours after feeding a dose of iron, a marked increase in ferritin takes place, reaching a maximum in about seven hours and declining after 2-5 days to normal. *One might postulate that the presence of ferritin was in some way connected with the presence of the mucosal bloc*, that is, only when ferritin in the mucosal cells was depleted would more ferrous iron be absorbed.

One may detect ferritin by the following simple procedure: Several centimeters of the duodenal mucosa of a guinea pig are scraped onto a slide and a few drops of 20 per cent $CdSO_4$ solution are mixed with this tissue. Then a cover slip is placed on it, and the liquid is permitted

to evaporate slowly overnight. Examination of the duodenal mucosa of the normal guinea pig in this manner reveals some tiny ferritin crystals. These crystals are quite rare and it is necessary to hunt for them with an oil immersion lens. However, if 10-20 mg. of ferrous iron are fed to a guinea pig and its duodenal mucosa is examined the next day, then several hundred ferritin crystals may be found on the slide. The highest concentration of ferritin is found in the duodenal region, less in the jejunum, and only traces in the ileum, large intestine and stomach.

It is a surprising fact that colorless crystals, i.e., of apoferritin, are not found in the mucosa. This protein crystallizes just as readily as it does when it has iron hydroxide micelles attached to it, i. e., apoferritin crystallizes as well as ferritin. If ferrous iron is fed, ferritin crystals can be demonstrated. This means that the feeding of iron in some way brings about an increase of a specific protein, apoferritin, to which the iron attaches in micelles of a particular kind of iron hydroxide, forming the compound, ferritin. A plausible way of looking at this phenomenon is to consider that apoferritin is constantly being synthesized and broken down. If iron hydroxide micelles attach to the apoferritin then the apoferritin is prevented from being broken down. Thus feeding of iron will lead to an accumulation of ferritin (Fig. 2).

Serum iron. Let us next consider how the serum iron reflects various aspects of iron metabolism and whether the serum iron level may be related to absorption of iron by the mucosal cells. A number of careful studies on this subject have come recently from the laboratories of M. M. Wintrobe[19] and C. V. Moore.[18] As has been noted before, the iron in the serum is transported by siderophilin, a β_1 globulin protein of molecular weight 90,000. Normally this protein is only one-third saturated with iron. If 10 mg. of ferrous iron are injected into the blood stream the serum iron can be raised from the normal value of about 100 γ per cent to about 300 γ per cent. Iron injected above this saturation level disappears very rapidly from the blood stream at the same time that toxic symptoms appear.[5, 22] This excess iron probably precipitates out and is filtered off by the lymph glands and by phagocytes.

The serum iron under normal conditions will reflect the level of iron available for hemoglobin synthesis.[23, 24] 1. The serum iron will be low in nutritional anemia (Table II) because there is too little iron

TABLE II. IRON ABSORPTION IN ANEMIAS

H = High L = Low N = Normal V = Very O = None	*Intestinal absorption of Fe*	*Fe Serum*	*Fe utilization for hemoglobin*	*Fe stores (in liver)*
I. *Lowered O_2 content in blood stream*				
A) Uncomplicated iron deficiency (1, 25)	H	L	H	L
B) Pernicious anemia (24, 25)	H	H	L	H
C) Refractory hypoplastic anemia (25)	H	H	L	H
D) Pyridoxine deficiency anemia (26)	H	H	L	V.H.
E) Hemolytic anemia (25)	H	H, L	H	H
F Copper deficiency anemia (23, 27, 28, 29)	H	L	L	H
II. *Anemia of infection or inflammation*				
A) Turpentine sterile abscess (30)	O	—	—	—
B) Chronic infection or inflammation (31-34)	—	L	L	H
III. *Hemochromatosis* (25)	H	H	N	V.H.

being absorbed by the mucosa or the iron stores in the form of ferritin have been too far depleted to supply the demand for hemoglobin synthesis.[1,25] 2. The serum iron will be high when hemoglobin synthesis is depressed as in the deficiency anemias, for example, pernicious anemia,[24,25] pyridoxine deficiency[26] and other hypoplastic anemias of unknown origin.[24,25] 3. Another important category of serum iron level is the one associated with chronic infection and inflammations where, in general, protein metabolism is adversly affected.[30-34] Here the serum iron is low because it is being swept into the storage depots; at the same time that the bone marrow activity and synthesis of hemoglobin are being depressed.

Is there a direct influence on the mucosal cells of a high or low serum iron level? If one increases the level of the serum iron to saturation by injecting ferrous gluconate just before feeding radio iron, there is no decrease in the rate of absorption of radio iron noticeable.[25] One may also inject iron-free siderophilin into the blood stream to increase

the level of this substance and this also has no effect on the rate of absorption of iron from the mucosal cells. *The serum iron is therefore to be regarded as a passive substance whose level does not influence appreciably the movement of iron from the mucosal cells into the blood stream.*

Oxygen and mucosal iron. As has been noted before, in uncomplicated iron deficiencies, say, due to hemorrhage, the mucosa responds to the anemic condition by permitting more iron to be absorbed (Table II). In this case the serum iron level is low. In anemia due to copper deficiency the serum iron level is also low.[23,27-29] However, in the anemias of pyridoxine deficiency, of pernicious anemia, and of hypoplastic anemia, the serum iron level is high. Now in all these anemias of whatever origin, and regardless of whether the serum iron level is low or high, the absorption of iron by the mucosa is found to be high.[25] It is clear that a factor on the blood stream side and connected with the anemic condition influences the movement of iron through the mucosal cell. The simplest view that might be considered is that the mucosal cell responds to the low oxygen supply brought about by the low content of hemoglobin in the blood stream. *Thus one might postulate that it is the low oxygen supply which is the factor which indirectly brings about the increase in the amount of iron moving through the mucosal cell into the blood stream.*

Just as the level of red cells in blood is governed by the O_2 supply to the marrow, so O_2 may be considered to affect the rate with which iron enters the blood stream. The intestinal mucosa has a very active aerobic metabolism.[35] It is conceivable that a decrease in oxygen supply would lead to a somewhat greater reducing tendency in these cells. A greater reducing tendency would lead to more ferric iron of ferritin being converted to ferrous iron and the ferrous iron would then diffuse into the blood stream. In a similar manner one might also picture the storage iron of ferritin in the liver, for example, as being made available more rapidly by a lowering of the oxygen supply. But the response to a lowered oxygen supply would depend on the normal metabolic level of the cell, the mucosal cells of the duodenum probably being more responsive to changes in oxygen than, say, the liver cells. Thus it has been shown experimentally[36,37] that iron absorbed by the intestinal mucosa is somewhat more readily available for heme synthesis in the bone marrow than is the storage iron in the liver.

Hypothesis on the regulation of iron by the mucosal cell. I have now finished presenting what appear to me to be the known pertinent facts about the absorption of iron and its regulation. The problem of iron absorption and regulation is a part of the larger problem of selective absorption and permeability. We are most strikingly reminded of our ignorance of such processes when we consider the functioning of the mammary gland, where the blood stream abuts one side of the cell and from the other side milk issues forth. It may seem foolish to even attempt an hypothesis of iron absorption and regulation at this stage of our knowledge. But I am going to make this attempt to outline a hypothesis on iron absorption and regulation for the purpose of summarizing and bringing together the facts into some coherent scheme. If such a hypothesis can suggest some correlations or new experiments it will have served a useful purpose.

Any proposed hypothesis must explain the following facts: 1. There is a one-way transfer of iron into the mucosal cell from the gut. 2. There is a resistance or bloc to iron absorption in the normal. 3. In the normal, after an acute anemia, there is a delay of about a week in responding to the anemia by an increased iron absorption. 4. After an iron feeding there is a rapid increase in ferritin (over a period of several hours) in the mucosal cells and then a rather slow decrease in ferritin (over a period of several days). 5. Apoferritin protein increases in the mucosal cells when iron is fed. 6. The chronically anemic condition brings about a greater absorption of iron.

The hypothesis is outlined in the scheme presented in Figure 2. We are going to postulate that a gradient of reduction exists in the cell, that portion of the mucosal cell closest to the lumen of the gut being less reducing for ferric iron, and that portion closest to the blood stream having a higher reducing ability for ferric iron. No direct evidence, however, exists for this assumption.

According to this hypothesis the iron would be absorbed in the ferrous form from the lumen of the gut into the mucosal cell. This passage is one-way. Once the iron enters the cell it never passes back again through this membrane. One possible explanation for this one-way movement might be that the ferrous iron once in the cell is rapidly oxidized and incorporated into ferritin. This would mean that the ferrous iron is perhaps enzymatically converted to the special 3-unpaired-electron type of ferric hydroxide polymer. Apoferritin protein mole-

cules may be postulated to be constantly turning over, i.e., being formed and broken down. When the ferric hydroxide polymers attach to the protein to form ferritin, the ferritin is stabilized so that the feeding of iron would lead to the accumulation of the iron hydroxide protein compound, ferritin.

Whether ferritin is directly connected with the mucosal bloc* or whether the presence of ferritin merely indicates that the cell is saturated with respect to some iron compound more closely allied with the mucosal bloc cannot be decided.

At the blood stream end of the cell, it is postulated, the ferric iron of ferritin is reduced to ferrous iron by the reducing mechanisms of the cell and enters the blood stream to be autoxidized to the ferric state and attached to siderophilin in the presence of CO_2. The concentration of siderophilin does not appear to affect the rate of movement of iron from the mucosal cell into the blood stream.

In uncomplicated iron deficiency anemia and other anemias, such as pernicious anemia, pyridoxine deficiency anemia, etc., the iron is absorbed more effectively into the blood stream. In these anemias the common feature appears to be a low oxygen supply in the tissues brought about by a low hemoglobin content of the blood stream. The low oxygen supply would lead to a relatively anaerobic atmosphere in the mucosal cell and might bring about a faster rate of reduction of ferric ferritin to ferrous iron and thus more ferrous iron might leave the cell.

According to this hypothesis, two mechanisms take part in regulating iron absorption in the mucosal cell. One is the mechanism of the mucosal bloc which is directly or indirectly connected with the ferritin content of the mucosal cells. The mucosal bloc may be regarded as a response to the cell being saturated with some iron compound. The second mechanism regulating iron absorption is postulated to be connected with the so-called "redox" level of the cell. The reducing systems are pictured as converting the ferric hydroxide micelles to ferrous iron, the ferrous iron then diffusing into the blood stream. In anemic conditions, the lower oxygen content in the mucosal cell would favor the reduction. Thus ferrous iron, absorbed from the gut might

* How can the presence of ferritin diminish the uptake of iron by the mucosal cell? One suggestion, purely hypothetical, is that some substance necessary for iron absorption in the cell might be removed from the cell and stored as ferritin. Only when the ferritin is broken down and the iron released would this substance be released.

stay in the ferrous form and move directly through this cell in this form. If conditions were normal, ferritin might be stored up and the bloc to further iron absorption would then be established.

IV. Hemochromatosis

Hemochromatosis is a disease characterized by an extensive deposition throughout the body of ferric hydroxide polymers in the form of brown iron-staining granules. These iron-staining granules are called hemosiderin granules. The deposition of hemosiderin may be especially marked in the liver and pancreas. The spleen and bone marrow contain relatively little of the pigment.[38, 39]

This iron-containing pigment is also accompanied by another pigment devoid of iron, called "hemofuscin." Hemofuscin may vary in color from gray to yellow to brown. It is very poorly characterized and may well represent several different substances, produced in the same or different cells as a result of abnormal cell metabolism.

Hemochromatosis is an interesting disease to students of iron metabolism since this disease appears to be a disorder connected with the absorption of abnormally large amounts of iron. Normally the total body iron may average 4-5 grams. In hemosiderosis the liver alone may contain 20 or more grams of iron. This accumulation of iron may be due to only a relatively slight increase in the efficiency of iron absorption as compared with the normal, since the excessive accumulation may be a process that goes on over a period of 20-30 years before it is detectable. As has been mentioned before, in order to accumulate some 30 gm. of iron over a period of 20 years as in cases of hemochromatosis, it would be necessary to accumulate about 3.5 mg. of iron per day, i.e., instead of the absorption of iron, from a meal containing 5 mg. Fe, being only 10 per cent efficient it would have to become 30 per cent efficient.

The first organ to be generally affected is the liver. This organ is the primary storehouse for iron, the iron being normally present as ferritin. If iron continues to be brought to the liver the following sequence of events may be postulated: The liver will form ferritin up to its capacity. The excess iron probably will continue to deposit on the already existing micelles increasing their size, and the links between micelles will tend to unite ferritin molecules to form clusters that eventually become large enough to be visible under the microscope (i.e.,

the hemosiderin granules). At the same time a portion of this iron may begin to spill over into other organs to be converted to ferritin and the excess finally to be deposited in the form of hemosiderin.

This process of iron deposition which is postulated to take place is probably normal in so far as the iron is converted to the normal 3-electron type of ferric hydroxide.[40] It has not been demonstrated that huge accumulations of iron in the form of ferritin and hemosiderin by themselves will bring about cell destruction with consequent fibrosis. Rather does it seem more reasonable to consider that iron which is not converted to the proper kind of ferric hydroxide may bring on cell destruction by precipitating proteins indiscriminately or perhaps by inactivating one or more specific enzymes.* If an acute inflammatory disease should arise in a subject whose liver is laden with ferritin and hemosiderin it is possible to conceive that the cells will not handle the iron properly at this time; and an excessive amount of an improper form of iron may then injure or irreversibly damage the cells. Another factor to be considered in hemochromatosis where ferritin is very high, is that injury to the liver may cause ferritin to escape into the blood stream leading to vasodepressor action of ferritin and its sequelae.

What factors may one postulate which might cause an excessive deposition of iron?

1. An accumulation of iron may be brought about as a normal response to a lowered oxygen supply. For example, in nutritional anemias such as in pyridoxine deficiency, pernicious anemia, copper deficiency, nicotinic acid deficiency, etc., the mucosal cells respond to the anemic condition in a normal manner, by permitting a greater than normal amount of iron to enter. In hemolytic anemia also, a greater amount of iron will enter than in the normal. Mallory[42] has claimed that a hemolytic anemia arising from chronic poisoning with copper might lead to hemochromatosis. Although iron derived from hemolysis would be found in especially large amounts in the spleen and bone marrow, it is possible that a redistribution of iron will occur over a period of years so that if a low grade hemolytic anemia had once been present and eventually disappeared one might not be able to recognize the fact by studying the tissues of the hemochromatosis case.

2. The hemochromatosis may be brought about as a result of ad-

* For example Racker and Krimsky[41] have called attention to the inhibitory action of iron on the important triose phosphate dehydrogenase enzyme.

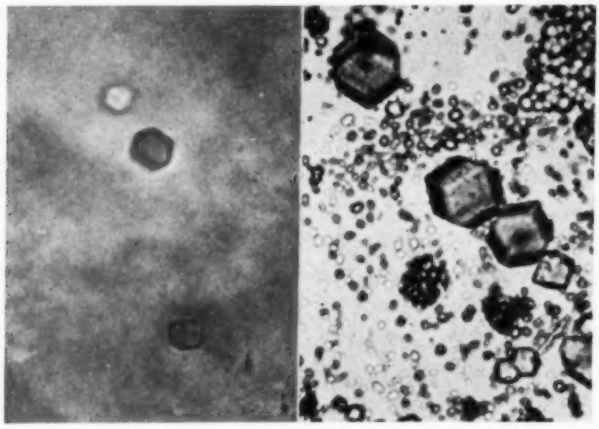


Fig. 3A Fig. 3B

A. Ferritin crystals from human duodenal mucosa of case of hemochromatosis.

B. Ferritin crystals from liver of a case of hemochromatosis brought on by a large number of transfusions.

ministering a large number of transfusions to a patient with hemolytic anemia. In one such case, after administration of about 200 blood transfusions the patient eventually died of hemochromatosis resulting from the iron liberated from the transfused blood. The liver was found to be extremely rich in iron and well developed crystals were obtained (Fig. 3B). The heart muscle also was found to contain ferritin. Schwartz and Blumenthal[39] have recently reviewed a number of such cases. They have noted that often the number of transfusions administered was too small to account for the high iron content of the tissues. This suggests that the anemic patients over a period of years had absorbed large amounts of iron in response to the anemic condition. The additional accumulation of iron from transfusions of blood seemed to bring out the symptoms of hemochromatosis.

3. Hemochromatosis may also be brought about by an inborn error of metabolism in the mucosal cells. Sheldon[38] has presented some evidence that this metabolic error may be inheritable. Lawrence[43] has described a family where the mother and two sons died of typical hemochromatosis, two sons had borderline cases, and two other sons

and three daughters were not affected.

The blood values in hemochromatosis are not abnormal, averaging about 4.1 million R.B.C. and 80 per cent hemoglobin. A diagnostic sign of hemochromatosis is the high serum iron, the siderophilin protein being saturated with iron. Overcoming certain objections to previous techniques, Dubach et al[25] in an excellent study, have described a case of hemochromatosis in which a test dose of 70 mg. of ferrous iron was fed by mouth. Of this test dose 20 per cent was retained by the hemochromatosis patient whereas the normal would retain 2 per cent or less of such a dose. This is the first direct evidence demonstrating that in hemochromatosis iron absorption is greater than normal.

Where does the abnormality lie in the mucosal cell which is responsible for hemochromatosis? We can examine this question only in the light of our limited knowledge. In terms of the hypothesis presented above (Fig. 2), two metabolic processes may govern the regulation of iron absorption by the mucosal cell. One is the sequence of reactions characterized by the mucosal bloc and related to ferritin turnover. The second is the process connected with the reducing ability of the cell.

If the processes related to the mucosal bloc are responsible it might be possible to show that some defect may be present that is connected with ferritin formation. We have examined the tissues of a case of typical hemochromatosis (i.e., cirrhosis of the liver, fibrosis of the myocardium) kindly provided us by Dr. R. West, with the idea that if the mucosal bloc were at fault then the mucosa might not form ferritin.[40] This idea proved to be incorrect. Ferritin crystals were obtained from the mucosa (Fig. 3A) and relatively huge amounts of ferritin were isolated from the liver. This indicates that no defect is present in this particular case of hemochromatosis, either in the formation of the iron hydroxide micelles which were of the characteristic 3-unpaired electron type, or in the enzymes connected with apoferritin and ferritin formation. The finding of ferritin in the mucosa makes it less likely that factors in the mucosal bloc may be involved.

It seems more likely that the metabolic error in hemochromatosis might be due to a slightly greater reducing tendency of the cell for iron. This would permit somewhat more iron to pass to the blood stream than in the normal. The greater reducing tendency or lower "redox" level might arise either by an increased effectiveness of the reducing enzymes or might be brought about by a decrease in the effectiveness

of the oxidizing enzymes.

To the practicing physician the practical question must be uppermost. What can be done for a patient with hemochromatosis? Little hope can be extended to a far-gone case except symptomatic treatment of cirrhosis, or the diabetes or heart involvement.

However, there would seem to be already a rational approach to the prevention of hemochromatosis. The hemochromatosis of the nutritional variety is probably extremely rare in this country. We are then concerned primarily with the hemochromatosis due to metabolic error.

1. Prevention and treatment depend on an early recognition of the development of this disease. A diagnostic sign is a rather high serum iron and an otherwise normal blood picture. If near relatives have had hemochromatosis there is all the more reason to be suspicious.

2. A diet might be considered which would limit iron absorption first by complexing the iron and secondly by keeping iron in the ferric state. A diet high in phosphates especially in phytic acid would render the ferric iron highly insoluble. One might consider increasing the oxidizing activity of the diet by adding vitamin K as the naphthoquinone. One might also decrease the intake of ascorbic acid in the food during meals to prevent the ferric iron of the food from being reduced.

3. One might consider the feasibility of making the potential hemochromatosis patient a regular blood donor since bleeding would be the only direct way of removing accumulations of iron from the body. In hemochromatosis developed to the stage of liver or pancreas involvement, however, this procedure might not be advisable.

4. An aim of the future, would be to study the metabolism of the mucosal cell in order to understand the factors governing the reducing mechanisms of the cell. Eventually one might attempt to control this activity by blocking one or another enzyme with some enzyme inhibitor or anti-metabolite.

In this talk I have attempted to summarize some of the aspects of our knowledge and hypotheses of iron metabolism and to relate them to the disease, hemochromatosis. The advances in our knowledge and changes in our viewpoint have been so great within the last 10-15 years that an important book on iron metabolism in relation to hemochromatosis by Sheldon, published in 1936, reads today like ancient history. I

hope that this rapid pace for advancing our knowledge of iron metabolism will continue and that this material which I have presented to you this afternoon will in turn shortly have the hollow ring of ancient history.

I should like to express my gratitude to Dr. L. Michaelis for his criticisms of this paper and his constant interest.

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THE PRESENT STATUS OF VITAMIN B_{12} IN PERNICIOUS ANEMIA*

EDWARD H. REISNER, JR.

WINTROBE states in his book, *Clinical Hematology*—"A confusing array of 'B vitamins' . . . have been shown to be related to blood formation." In the past pyridoxine, riboflavin, nicotinic acid and folic acid have been shown to play a role in hematopoiesis, and now vitamin B_{12} has been added to that group. It is the purpose of this paper to review briefly the steps leading up to the discovery of vitamin B_{12}, to discuss some of the facts we now know about it, and to show you the records of cases treated with it by Randolph West and myself.

Ever since the discovery of the effect of liver in pernicious anemia, successively more potent extracts of liver have been produced in the effort to isolate the active antianemic principle. These attempts were retarded by the necessity to test the potency of liver fractions by tedious clinical trial methods. With the discovery of folic acid a new technique of bioassay was developed, depending on the fact that certain bacteria required folic acid to achieve optimum growth. In 1947 Shorb[1] reported that the growth requirements of Lactobacillus lactis Dorner for liver extracts paralleled their antianemic potency. This provided an in vitro method of testing the potency of such extracts and expedited the research culminating in the isolation from liver last year by Rickes and his associates[2] of some reddish crystals, designated vitamin B_{12}. Simultaneously, the same substance was isolated by English investigators under Smith,[3] using the traditional clinical test methods.

Vitamin B_{12} is a red crystalline substance with a molecular weight of 1630.[4] It contains cobalt, nitrogen and phosphorus but no sulphur.[5] It is extracted from liver in exceedingly small amounts and can also be obtained from a variety of sources in nature, including the manure of cows, chicks and other species[6] and has recently been reported to be found in the liquor produced in the production of streptomycin. Its potency in the treatment of pernicious anemia was reported by West[7]

* From the Fourth Medical Division (New York University), Bellevue Hospital, N. Y. Given February 15, 1949 before the Section on Medicine of The New York Academy of Medicine.

and confirmed shortly by Spies[8] who also found it effective in sprue and nutritional macrocytic anemia. Its outstanding feature was its potency, doses as small as 4 micrograms being adequate to produce a maximal reticulocyte response.

The first patient treated was a 66 year old white female with pernicious anemia, a patient on the wards of the Kings County Hospital, made available to Dr. West through the courtesy of William Dock. Following a single injection of 150 micrograms a maximal reticulocyte response of 27 per cent was observed and the blood count rose to normal and is still over four million, ten months later, with no further therapy.

Having established the efficacy of the material, a series of trials was instituted to establish the limits of potency, using patients on the First and Fourth Medical Divisions of Bellevue Hospital. It was found that a single injection of as little as 3 to 6 micrograms produced a maximal reticulocyte response, and a total dose of 56 micrograms in one patient effected complete hematologic remission, which was maintained for four months before relapse, with no additional therapy. A dose of one microgram a day was found to give a maximal reticulocyte response and restore the blood count to normal. A daily injection of 1/10 of a microgram was ineffective, but remission occurred when this was increased to ¾ of a microgram daily. One-half a microgram daily gave a submaximal reticulocyte response. From these experiments the unit potency of B_{12} was fixed at approximately one unit per microgram, a unit being defined as the amount of antianemic substance required daily to effect and maintain hematologic remission in a patient with pernicious anemia.

The next problem to investigate was the effect of B_{12} on the neurologic lesions of combined sclerosis. One of the Bellevue patients had exhibited early cord lesions at the onset of treatment with one gamma a day, and at the end of fifty-three days of treatment had a negative neurological examination. Four patients with more severe neurological disease were treated at the Columbia-Presbyterian Medical Center by Dr. West. The dose employed here was 25 micrograms a week. There was marked improvement in ability to walk, and gain in subjective and objective motor strength and coördination. The neurologic signs such as Babinski and Romberg signs tended to diminish or disappear. Vibratory sense on the other hand improved slowly and only slightly. These

patients have now been followed for eight months, and on 25 micrograms a week have maintained their improvement and shown no sign of relapse. In general the neurological results of treatment with B_{12} seem to be as satisfactory as those that might have been obtained with vigorous liver therapy.

Meanwhile other workers have been investigating various aspects of B_{12} treatment and reporting their findings which will be briefly summarized. Hall[9] reported beginning disappearance of megaloblasts from the bone marrow as early as 18 hours after the injection of B_{12}. Bethell[10] demonstrated that a substance probably the same as B_{12} was excreted in the stools of patients with pernicious anemia in relapse, and that this substance when extracted from their stools and given parenterally, caused remission. Berk et al[11] reported that when B_{12} was given by mouth it was ineffective unless gastric juice was given with it, an observation confirmed by Hall.[12] From these facts it has been suggested that the role of the intrinsic factor of Castle may be to promote the absorption of B_{12} from the gut, and B_{12} has been suggested to be the extrinsic factor.

The question will at once be asked about the relationship of B_{12} to folic acid. Bethell[10] reported that the response to B_{12} was inhibited by folic acid antagonists. Sturgis[13] has reported that B_{12} is ineffective in the so-called pernicious anemia of pregnancy, which is cured by folic acid, and Luhby reports that B_{12} is likewise ineffective in the treatment of acute megaloblastic anemia of infancy, which also responds to folic acid. Folic acid and B_{12} probably operate at different levels in the process of hematopoiesis.

It has been shown that thymine in a ratio of several thousand to one can replace folic acid in the growth requirements of Streptococcus lactis R[14] and Lactobacillus casei.[15] Spies[16] gave thymine in similarly large doses to patients with pernicious anemia and sprue, and obtained restoration of the blood count. Like folic acid, however, thymine had no effect on the neurologic lesions in these conditions.[17] Wright[18] has shown that the growth requirements of L. lactis can be met by substituting thymidine for B_{12} in a ratio of 10,000 to 1. Thymidine is the desoxyriboside of thymine and from the foregoing evidence it is tempting to postulate that the role of folic acid is to act as a coenzyme in the formation of thymine, which is then converted to its desoxyriboside by the enzymatic action of B_{12}. We have attempted to treat patients

with pernicious anemia with thymidine in dosage up to 150 micrograms, without notable effect. While such an hypothesis is attractive it is probably oversimplified and fails to take into account a wealth of accumulated evidence concerning the role in hematopoiesis of other factors, including xanthopterin,[19] and other pterins. Most recently a highly potent hematopoietic substance called B_{14} [20] has been announced, but this claim is awaiting confirmation at present.

Finally, the presence of cobalt in B_{12} is certain to excite interest because of the fact that this element has been implicated for many years in the production of experimental polycythemia.[21] With the aid of its isotopes it is now possible to learn more about the fate of cobalt in the body,[22] and the presence of this inorganic ion in B_{12} may ultimately be of great help in studying the formation and role of this vitamin. Two of our patients were treated by West with cobaltous chloride with no effect, before they responded to B_{12}.

To summarize, it would appear that vitamin B_{12} is identical with the long sought anti-pernicious anemia fraction of liver. It has an antianemic potency of one unit per microgram of crystalline substance, and brings about complete hematologic remission, and improvement of the neurological lesions comparable to that obtainable with liver. In the light of recent observations it would appear that our twenty-year-old concept of extrinsic and intrinsic factors in pernicious anemia may have to be modified, and to end in the vein in which I started, the "array of B-vitamins" has been extended, and the confusion, which I have tried tonight to dispel to some extent, is still very dense.

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PAIN IN THE LUMBOSACRAL REGION*

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BACKACHE both with and without radiation of pain into the lower extremities is an exceedingly common complaint. It impairs the efficiency of thousands of manual workers, curtails the activities of some seeking physical recreation and accounts for the acid disposition of many an otherwise pleasant housewife. Last year alone, some 2200 patients with pain in the lower back were considered by the officials of the New York State Industrial Commission and other thousands consulted orthopedic surgeons, neurosurgeons, osteopaths and chiropractors. The fact that many of these wander from doctor to doctor as well as to members of legally unrecognized healing cults clearly indicates that we are as yet incapable of determining the nature of many of the lesions that produce pain in the lower part of the back and that there is no specific therapy for a large number of these sufferers. To label such complaints as sacroiliac strain, arthritis, or one of a host of other appelations that mean nothing and certainly bring little, if any, help to the patient, tends to terminate investigation that may eventually lead to a correct diagnosis. It seems best that the "book be kept open" for those with backache of undetermined cause, and gradually, as in the case of herniated intervertebral disc, other clinical syndromes may be delineated from the stock pile of patients with low back pain.

It is therefore appropriate to first consider the syndromes characterized by pain limited to the lower back. In most instances the pain is relatively sudden in onset, usually precipitated by physical exertion and often aggravated by any movement of the lower vertebral column. Frequently the patient states that at the outset "something tore loose in my back" or "something slipped out of place." In others, the initial discomfort is relatively mild but after a night's rest they are unable to get out of bed or, once up, are unable to dress themselves. Those so

* Read November 5, 1948 in Friday Afternoon Lecture Series at The New York Academy of Medicine.

afflicted walk with a cautious gait and many have a slight limp. The trunk may be tilted forward or laterally and the upper extremities may be carried in slight abduction to aid in balance. Difficulty in arising from a sitting position is common and to retrieve an object from the floor is impossible. The pain is described as a continuous boring ache deep in the middle of or to one or the other side of the lumbar region of the back. During periods of exacerbation the pain tends to spread over the buttocks and into the upper thighs. Lying supine on a hard surface usually gives considerable relief. Examination is usually not very enlightening. The normal lumbar lordosis is obliterated in some, more often the sacrospinalis muscles are unduly prominent due to continuous spasm. These paravertebral muscles on one side may be more spastic resulting in mild to moderate scoliosis. Flexion, extension and lateral mobility of the trunk are limited, and commonly painful. By mild percussion one may demonstrate a well localized area of tenderness. In the supine position the pain in the back is aggravated by flexing the thigh with the leg in the extended position and this is usually more evident on one side. The tendon reflexes of the lower extremities are seldom changed and there is no alteration in the cutaneous sensibilities except in some patients there may be increased skin sensitiveness over the lower lumbar area. Early in the course of the disease roentgen-ray examination is seldom helpful. The abnormal posture of the spine may be demonstrable, an intervertebral space mildly narrowed, or in the older age group an overgrowth of bone at the vertebral margins may be observed. Narrowing of an intervertebral space as well as so-called hypertrophic arthropathy of the spine are too commonly observed in patients without pain for one to assign these as common causes for the acute disorder under consideration. Rarely, the protein content of the cerebrospinal fluid is moderately elevated and this without clinical evidence that the intradural structures are implicated. When all the available evidence is evaluated one is often forced to conclude that the patient has "pain in the lower back . . . cause unknown." There are, however, a few patients with this syndrome who have changes in the vertebrae demonstrable by roentgen-ray examination. One may arrive at an accurate conclusion regarding these and carry out appropriate definitive therapy.

For those of unknown etiology, conservative palliative measures are indicated followed by repeated observation including roentgen-ray

examinations of the involved part. The spasm of the paravertebral musculature which seems to be nature's attempt to immobilize the spine points the way for palliation. By and large, the more complete the immobilization the more effective the relief from pain. It is evident that complete immobilization is impossible in ambulatory patients. Effective limitation of movement of the trunk may be obtained by a 14 to 16 inch corset laced snugly about the lower thorax and pelvis. Other patients may be made more comfortable by a plaster of paris jacket and for still others a period of complete bed-rest is necessary. There are a few who will eventually require internal fixation by fusing the distal vertebral segments to the pelvis. It is always most important to remember that in the instances in which the causative factor for the pain has never been determined, periodic re-evaluation of the problem as if one had never seen the patient before will occasionally disclose the reason for the continued pain. Certainly surgical intervention should seldom be carried out for pain limited to the lower back unless the diagnosis is obvious and such therapy is clearly indicated.

Pain in the Back With Radiating Pain Into One Lower Extremity

Not infrequently a pathological process in the lower lumbar region will result in pain limited to this area and later as a nerve root or roots become implicated the pain begins to radiate into one lower extremity. A long list of such lesions could be compiled, however, the ones commonly encountered are: fractures of vertebra, herniated intervertebral disc, spinal epidural or metastatic tumors (later involving both lowers), spinal epidural abscess, osteomyelitis, paravertebral metastatic tumor.

Of these, the outstanding examples are herniation of a part of an intervertebral disc and carcinomatous metastasis of the spinal column. Patients with the syndrome of herniated disc are more frequently encountered in office practice whereas in large municipal hospitals, carcinoma that has metastasized to the spine is an exceedingly common cause of "sciatica." The early symptoms of most patients with herniated disc are referable to the lower back. After several bouts of pain in the lower back, usually precipitated by bending or lifting and lasting from a few days to weeks, the patient begins to have pain in one buttock, along the posterolateral aspect of the thigh, in the lateral aspect of the leg and often terminating in the posterior part of the ankle. In some

the pain extends into the lateral aspect of the foot involving the fourth and little toes; in others the pain extends across the dorsum of the foot into the great toe. Pain in the back, buttocks, thigh, leg and outer side of the foot suggests a herniation of the fifth intervertebral disc whereas pain as above described radiating into the big toe suggests a herniation of a part of the fourth lumbar intervertebral disc. In addition, herniations at the fourth interspace often cause pain in the groin and this, if present, is usually experienced before the pain has extended into the lower extremity. It therefore may be said that in most instances the symptoms and abnormal physical signs that a patient should have before he is subjected to operation are: recurring attacks of pain or stiffness in the lower back with radiation of pain along the posterolateral thigh, "through" the knee into the calf or lateral aspect of the leg; numbish feeling in the lateral aspect of the leg; relative freedom from pain between attacks; moderate to marked fixation of the lumbar spine by muscle spasm; mild scoliosis (usually the lumbar spine is tilted away from the side of the lesion but under certain circumstances may be tilted toward the lesion); tenderness to percussion over the site of suspected herniation; mild diminution to absence of the ankle jerk; slight atrophy of the involved leg if symptoms are long-standing; hypalgesia of the anterolateral leg and medial dorsal part of the foot in lesions at the L_4 level; hypalgesia of the lateral leg and lateral one-third of the foot in herniations at the L_5 level. Variations of symptoms will be encountered with herniations at these common sites for the lesion (L_4 and L_5 intervertebral levels) and, as would be expected, the distribution of pain associated with the uncommon herniations at the L_1, L_2 and L_3 levels will be in accord with the spinal nerve root implicated. One should not rely on narrowing of an intervertebral disc as disclosed on plain x-ray films as an indication of the presence of a herniation at the level of the narrowing. Narrowing of the disc may be associated with dorsolateral herniation of a part of the disc; however, in the majority of instances this is not the case. Furthermore, one should not subject a patient to myelography unless there are unusual symptoms and physical signs that cannot be interpreted without the aid of this examination. A correct diagnosis is possible on clinical grounds alone in ninety per cent of patients with herniations of a part of the fourth or fifth intervertebral disc in need of surgical therapy. If there is doubt regarding the cause of the pain, be conservative. This implies relative immobilization of the

lumbar spine either by bed-rest or external appliances and the administration of analgesics for a few days to bring the acute pain under control. Paravertebral injection of a solution of novocaine will at times give some relief but the results are too uncertain for the method to be generally applicable.

Any pathological process involving a single nerve root (L_5 and S_1 in particular) may give rise to symptoms and signs comparable to those of herniated disc. The most common lesion, however, causing pain in the back and one lower extremity to be differentiated from herniated disc is metastatic carcinoma. Perineural extension of the cancer into the region of the fourth or fifth lumbar intervertebral foramen may result in symptoms simulating herniated disc. Pain due to cancer of the distal lumbar spine is usually continuous: mild, early in the course of the disease, but gradually becoming unbearable. Even the most severe pain caused by herniated disc is usually somewhat relieved by the recumbent position whereas the pain of cancer is seldom altered by change of posture. This is particularly true regarding the pain in the back. As a rule, by the time metastatic carcinoma of the spine has advanced sufficiently to cause pain in one or both lower extremities, destructive changes in the vertebral bodies are demonstrable by roentgen-ray examination. Not infrequently patients are encountered complaining of pain in the back with radiation along the anterior aspect of the thigh, numbish feeling in this area and diminution to absence of the knee jerk. As in the other syndromes, a variety of causative factors may be disclosed, however, this particular symptom-complex is most commonly the result of paravertebral metastasis or an extension into the paravertebral region of a regional malignant process. The spinal nerves are often implicated paravertebrally long before the vertebral bodies are involved. Not infrequently it is necessary to perform myelography to differentiate a lesion so placed from a neoplasm within the spinal canal.

Pain in the Lower Back With Radiating Pain Into Both Lower Extremities

Except in examples of accidental trauma and in some instances of inflammatory lesions of the cauda equina, one seldom encounters patients who have had pain appearing simultaneously in both lower extremities. Regardless of the nature of the pathological process, those patients who eventually have pain in the back and both lower extremi-

ties usually have the initial pain limited to the back; later in the course of the disease there is pain in one thigh or leg and finally radiation of pain into both lower extremities. The following are some of the lesions that produce this syndrome: fracture of vertebra, intradural tumor, metastatic tumor of vertebra, herniated disc (occasionally), vertebral osteomyelitis (pyogenic, tbc.), postmeningitic adhesions (cauda equina).

New growths, either metastatic or primary, are the leading causes for radicular pain involving both lower extremities. Only rarely may large herniations of an intervertebral disc produce pain in both legs. The primary neoplasms are most often located in the vertebral canal, some epidural, others within the dural envelope. Those outside the dura but within the spinal canal are frequently extensions from a malignant process elsewhere, the lymphoblastomas being the most common. Those within the dura are for the most part gliomatous, arising from the filum terminale; however, a more benign tumor belonging to the meningioma group is occasionally encountered in the lumbar region. A more elaborate description of the symptoms and signs resulting from compression of the cauda equina seems inappropriate here. Suffice it to say that all patients with pain in the back and both lower extremities should be studied critically for a malignant process that has metastasized to the distal lumbar spine. In the absence of evidence of such a process in the vertebra, myelography may be indicated. Many of the primary neoplasms that grow within the dura lend themselves to total removal, however, it is seldom that those located in the epidural space can be cured by surgical therapy. The continued growth of these as well as the metastatic tumors of the vertebral bodies often produce intractable pain. Temporary respite may be obtained by intensive roentgen-ray therapy. Sooner or later it becomes necessary either to interrupt the pain-carrying pathways of the spinal cord by anterolateral cordotomy or, in some instances, to perform prefrontal leucotomy. Formerly only the spinal operation was available and this never was too satisfactory due to the fact that when performed on both sides of the cord, undesirable and troublesome disturbances in the urinary and anal sphincters ensued. Therefore this operation is being replaced by the prefrontal lobotomy. It is thought by some that appropriate division of the fiber pathways in one frontal lobe suffices, thereby obviating the intellectual blunting that may follow the operation when performed bilaterally. In our experience the one-sided procedure does not give satisfactory

results and in all the patients in whom this was carried out, the complaints of severe pain returned, necessitating the section on the other side. Recently it has been stated that the operation called topectomy now being investigated by the group at the New York Neurological Institute has been effectively employed for the control of pain. It is too soon to evaluate this procedure.

Finally, in all situations that have been listed in the three categories under consideration, surgical therapy should be employed only when the evidence available indicates a lesion amenable to such therapy. All patients with pathophysiological disturbance of unknown pathogenesis should be temporarily treated empirically but critically reviewed at intervals in an attempt to arrive at a correct diagnosis.

CLINICAL RESEARCH MEETING

Arranged by the Committee on Medical Education

JUNE 1 and 2, 1949

An Evaluation of the Mumps Skin-Test in Pediatric Practice

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The intradermal inoculation of killed mumps virus was introduced in 1945 by Enders, Cohen and Kane as a measure of resistance to mumps infection. The test gives a tuberculin-like reaction. Those who have had clinical or inapparent mumps previously react positively.

This test was employed during 1948-1949 in the course of a study of the epidemiology of mumps in household groups such as are seen in private pediatric practice in New York City. The antigen was formalin killed egg grown virus partially purified by centrifugation. Forty households were selected in which at least one member was ill with the disease. Skin-tests were performed on 135 individuals, 26 of whom were first cases and 109 of whom were contacts. Among the contacts there were 80 adults and 29 children.

The reactions, especially among the children, were often difficult to read. The erythema was sometimes faint and its edges poorly differentiated. All of the first cases gave negative reactions during acute illness. When 6 were retested several months later 5 had become clearly positive and one questionably so. Among the contacts there were 60 positive and 49 negative skin-tests. Only one half of the positive reactors definitely recalled having had the disease.

Within the following 3 weeks, 12 of the contacts became ill with clinical mumps. Only one of these contact cases was among the positive reactors and 11 were among those with negative skin-tests. Since the 49 negative reactors consisted of 22 adults and 27 children, it is significant that 10 of the secondary cases occurred among the children.

It is concluded that although the test is useful in determining susceptibility to mumps, a positive skin reaction is not the sole indicator of resistance.

* Aided by a grant from the Sara Welt Foundation.

Hypermetabolism Without Hyperthyroidism

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It is an accepted fact that there are clinical conditions in which the basal metabolic rate is elevated in spite of the fact that none of the features of hyperthyroidism is present. Clinicians have known for a long time that many of the patients suffering from hypertension, polycythemia, leukemia, malignant lymphomas, generalized or local-

ized malignant growths, Paget's disease of the bones, and other diseases frequently present an elevation of the basal metabolic rate. It is also well recognized that these patients do not present the classical signs of hyperthyroidism and the pathological changes in the thyroid gland do not suggest the typical hyperplasia and other alterations of Graves' disease.

We have attempted to study thyroid function in this group of patients with elevated basal metabolic rates by determining the level of the non-dialyzable (protein-bound) blood iodine and the urinary excretion of tracer doses of I 131.

Both of these methods show constant changes in hyperthyroidism. In this disease the "hormone" iodine is consistently elevated in the blood and the urinary excretion of tracer doses of I 131 is reduced due to the increased avidity of the thyroid gland for iodine.

In a series of over 100 consecutive patients suffering from the diseases outlined above whose metabolic rates were elevated and who presented no clinical signs of Graves' disease, we determined the blood iodine levels in all and the I 131 excretion in some. Without exception, the blood iodine levels and I 131 excretion were normal in spite of very marked elevations of the basal metabolic rate.

Summary: In hypermetabolic states not due to hyperthyroidism, the function of the thyroid gland is normal as measured by the level of the protein-bound blood iodine and the urinary excretion of I 131. The increased metabolism in these disorders is apparently not mediated through the thyroid gland.

The Association of Interatrial Septal Defect and Anomalies of the Osseous System

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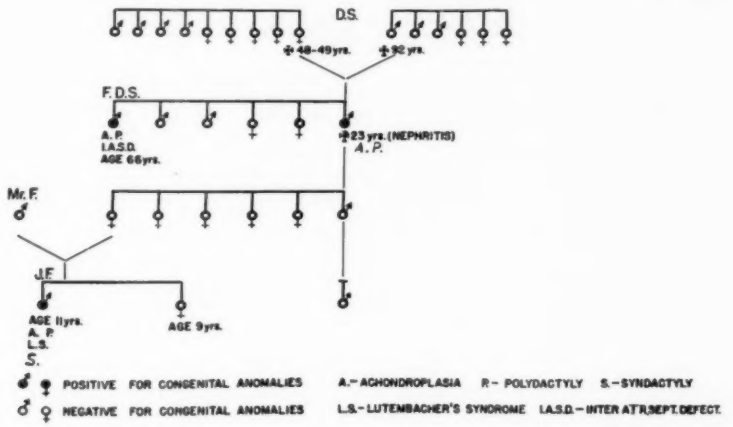
The association of congenital defects of the heart with those of the locomotor system is relatively rare. Arachnodactyly has been reported in combination with interatrial septal defect and with congenital aneurysm of the aorta. Achondroplasia in association with patent ductus arteriosus has been mentioned once in the literature. We have observed several cases of congenital cardiac defects in patients with rare anomalies of the osseous system. The congenital cardiac anomaly encountered was invariably an interatrial septal defect either alone or in association with a congenital mitral stenosis. Five illustrative examples of the association of such a congenital cardiac anomaly with various abnormalities of the osseous system are herewith reported.

In the first 2 patients (grandfather and grandson) the family history is of particular interest, as depicted in the accompanying genealogical chart. The grandfather (case 1) is an achondroplasic dwarf, with polydactyly of the hands, interatrial septal defect, hypertension, coronary artery disease and congestive heart failure.

The grandson (case 2) now an 11-year boy presents the following congenital defects: achondroplasia, polydactyly, syndactyly and interatrial septal defect with congenital mitral stenosis (Lutembacher's syndrome).

In the third case, no congenital stigmata are known to exist in the family. He is a 12-year old boy. His bony deformities consist of congenital synostosis of several cervical vertebrae (Klippel-Feil syndrome), bilateral multiple anomalies of the carpal bones, cervical ribs and subluxation of the right thumb. His cardiac anomaly is an interatrial septal defect.



Patient 4 is a 21-year old male whose parents are first cousins, but free of congenital anomalies. The patient has polydactyly and so has his sister, her only daughter, and his first cousin's daughter. None of the members of his family have heart disease. The patient himself is only 4 feet 9½ inches tall, weighs 90¾ lbs. and is of proportionate build, with almost no pubic, facial or axillary hair. Polydactylism and webbing of the second and third toes of both feet are present. His cardiac anomaly is Lutembacher's syndrome. He is a hypophyseal dwarf.

The fifth patient, a 6-year old girl, was the first member of her family to be afflicted with multiple congenital anomalies. She has polyphalangism of both thumbs associated with bilateral absence of opponens action of these digits. The cardiac lesion was a Lutembacher's syndrome.

We find that the congenital cardiac lesion when associated with congenital bony defect is usually an interatrial septal defect, with or without congenital mitral stenosis. The familial occurrence of the osseous anomalies in 3 patients indicates that the cause can hardly be ascribed to an intra-uterine fault. The presence of an interatrial septal defect in all of the 5 cases suggests that the lesion is rather of genetic than of intra-uterine origin.

Simultaneous Left and Right-Sided Intracardiac Electrocardiography in Man: Right Bundle Branch Block

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Catheterization of the right cardiac chambers and pulmonary artery has been widely applied to the study of cardiac dynamics and for the analyses of right sided intracardiac potentials. Retrograde catheterization of the aorta from the femoral, brachial, or axillary artery has been employed for direct aortography. As yet, retrograde left cavity catheterization for the study of hemodynamics or cavity potentials in humans has not been reported. During our intensive study of cavity surface, esophageal, chest and extremity potentials in man, we thought that the simultaneous recording of

right and left sided cavity potentials would be important. Patients with right bundle branch block permit particularly well the analyses of the initial ventricular deflection in the left ventricular cavity without interference from right ventricular activity.

The retrograde catheterizations of the left ventricle were carried out from either common carotid artery. The radio-opaque catheter was either a No. 3.5 French fitting through a No. 16 gauge needle or a No. 5.0 French fitting through a No. 13 gauge needle. With direct surgical exposure as done for cerebral arteriography, the needle with the catheter was inserted into the common carotid artery and directed against the blood stream towards the aortic valve. The catheter was positioned under fluoroscopic control with the needle remaining in the carotid artery. There is less trauma to the arterial wall if the needle remains in position and serves as a sleeve for the catheter. Retrograde passage through the aortic valve was very difficult because of the high forward pressure. It was best accomplished by allowing the catheter to form a coil. The catheter for recording the right cavity potentials was introduced into an antecubital vein of either arm. Spot films were taken whenever electrocardiograms were recorded.

Because this study was of investigative nature, patients with advanced metastatic malignancy were chosen as subjects.

The electrocardiogram in the case of the right bundle branch block to be demonstrated shows the following:

Left ventricular cavity: deep QS pattern with a terminal second QS shaped wave and positive T-wave. The stimulus traversing the septum from left to right reaches the right ventricular cavity in 0.025 sec.

Right ventricular cavity: the initial R-wave reaches its peak in 0.02 sec. and the second R wave in 0.08 sec., evidence of marked delay of right ventricular stimulation. The T wave is inverted.

These findings are identical with those obtained by Wilson et al. in animals. In normal individuals, right ventricular cavity leads often but not invariably show an initial R-wave. In this case of right bundle branch block the left ventricular cavity lead shows a QS wave. It therefore appears that in humans as in canines the stimulus to the ventricles reaches the left portion of the septum first and only then spreads to the right side.

Studies are in progress to supplement these studies with left ventricular catheterizations via the pulmonic veins which are exposed during pneumonectomies.

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*Anomalous Atrioventricular Excitation Produced By Catheterization of the Normal Human Heart**

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In the course of a comprehensive study of the intracardiac potentials developed during electrical activity of the human heart we have on two occasions, while manipulating the catheter, seen what appeared to be anomalous atrioventricular excitation (preexcitation) in normal subjects. In both instances the abnormal rhythm occurred while the catheter was being moved in the heart or just after movement had ceased. On one occasion there was some aberration of the ventricular complex in the intracardiac record which was not reflected at all in the simultaneously recorded external lead. On other occasions, ordinary premature systoles of ventricular origin occurred.

The data may be interpreted in one of three ways: 1) the catheter increased the

rhythmicity of a center in the ventricular muscle which then fortuitously discharged at a rate similar to, but preceding, impulses from the sinoatrial node; 2) the catheter itself, extending as it does through the right atrium, was moved by contraction of this chamber to a sufficient degree to make more distal parts of it in the ventricle stimulate the latter prematurely, probably in the region of the upper right side of the interventricular septum; 3) the catheter increased the irritability of a ventricular center which was then discharged prematurely by atrial systole, either electrical or mechanical.

By indirect reasoning, the third interpretation is believed to be correct.

In the past, two mechanisms have been advanced to explain the spontaneous occurrence of the type of electrocardiogram under discussion. One of these is anatomical, the other physiological. The former ascribes anomalous atrioventricular excitation to the existence of one of several anomalous pathways of conduction between the atria and the ventricles. A fallacy of the anatomical concept has been to ascribe a function of conduction to a tract simply because it has been found to exist at necropsy in a patient known to have shown the abnormal electrocardiogram during life. Adequate attention does not seem to have been paid to control observations; it is not known what number of subjects will display such tracts at necropsy who never had the abnormal electrocardiogram during life.

The data presented do not disprove the anatomical concept but do revive interest in a possible physiological approach to a final understanding of the exact mechanism in anomalous atrioventricular excitation.

Ergotamine Tartrate and the "2-Step" Exercise Electrocardiogram in Functional Heart Disturbance and in Organic Heart Disease

LEON PORDY, JOSEPH KOLKER, MORTIMER J. BLUMENTHAL and ARTHUR M. MASTER

Patients with severe neurotic complaints, including chest pain, may exhibit pronounced RS-T depressions and T-wave inversions in the "2-step" exercise electrocardiogram and in the 10 per cent oxygen test, indistinguishable from those found in organic heart disease. Because of the obvious clinical implications stemming from these facts, we have attempted to elucidate the mechanism of the electrocardiographic aberrations in psychoneurotic subjects following the "2-step" exercise test and to offer a means for the differential diagnosis of organic and functional heart disturbance.

The "2-step" exercise tolerance test was performed in ten patients with signs and symptoms referable to the heart before and after the intravenous administration of ergotamine tartrate. In the five cases which were classified as functional the "2-step" exercise test was positive before ergotamine but negative after its administration. In the five cases known to be affected with organic coronary artery disease the "2-step" test was positive both before and after the ergotamine injection. In other words, in the patients with functional heart disturbance there was consistent prevention of electrocardiographic abnormalities following exercise by ergotamine, whereas in patients with organic coronary artery disease the administration of ergotamine did not alter the abnormalities which appeared after exercise.

Ergotamine was administered intravenously in ⅓ to ½ mg. doses. The effects became marked in 15 to 30 minutes and lasted one hour or more. A drop in pulse rate of 20 to 30 beats per minute was common along with a rise in blood pressure of 20 to 40 mm. of mercury. Elevation of T-waves was observed.

The exact mechanism of alteration of the RS-T segments and T-waves of patients with functional disorder is unresolved. Lability of the autonomic nervous system, diminution in coronary blood flow or direct nervous effects on cardiac metabolism have been suggested.

It has been accepted that ergotamine is adrenolytic and sympatholytic in the doses employed in experimental animals, usually of the order of 0.05 to 0.1 mg/kilo body weight. There is no definite evidence that in the dosages employed clinically in man (0.5 mg.) it possesses similar properties. The complicated effects of ergotamine tartrate on the heart, including its poorly understood nervous system effects, and the vasoconstrictive action on the coronary vessels preclude any simple explanation of its effects on the electrocardiogram of neurotic patients. Despite the theoretic uncertainties touched upon above, its effects are quite definite in the prevention of abnormalities in the electrocardiogram of patients with functional heart disorders.

The side reactions to ergotamine tartrate given intravenously were nausea, vomiting, and in two of the patients with coronary artery disease, attacks of angina pectoris. The routine clinical use of ergotamine is contraindicated in such cases because of its anginal provoking properties.

Further studies on the relationship of the vegetative nervous system and the electrocardiogram with the newer autonomic drugs may elucidate some of the problems raised by this study and provide accurate clinical methods for distinguishing functional from organic heart disease.

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*Effect of An Adrenolytic Compound, Dihydroergocornine, on Epinephrine-Induced Stimulation of the Adrenal Cortex**

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Epinephrine has been shown by Vogt to be a physiological stimulus to the production of adrenal cortical hormone, presumably mediated via the adrenocorticotrophic hormone of the anterior pituitary (Long). Because of the possible bearing of this mechanism on the adaptation syndrome of Selye, and also on the question of neurohormonal interrelationships in hypertension it was thought of interest to investigate whether the stimulation of the adrenal cortex by epinephrine could be inhibited by adrenolytic compounds. Dihydroergocornine has been shown to possess potent adrenolytic and sympathicolytic actions, i.e., inhibition of excitatory epinephrine effect on rabbit uterus, inhibition of epinephrine induced hyperglycemia, inhibition of relaxing effect of epinephrine on intestine (Rothlin), diminution of pressor response, tachycardia and mydriasis following epinephrine (Freis, Stanton and Wilkins).

As an index of adrenal cortical stimulation the fall in eosinophiles and lymphocytes produced by epinephrine was studied, employing the Laragh modification of the test described by Thorn and co-workers. Following control eosinophile and lymphocyte counts 0.5 mgm. of epinephrine was given subcutaneously to eight subjects, twenty minutes after intravenous injection of 0.5 mgm. of dihydroergocornine. The response was compared with control administration of epinephrine alone two to four days later. Following epinephrine alone an average reduction in eosinophiles of 54.6 per cent from an initial count of 194/mm^3 occurred, whereas when epinephrine was given following dihydroergocornine the average decrease in eosinophiles was 56.5 per cent from an initial count of 201/mm^3. Reduction in the lymphocyte count following epinephrine

alone was 29 per cent; following epinephrine and dihydroergocornine an average fall of 53 per cent occurred. Control lymphocyte counts were found to be more variable than eosinophile counts, and the eosinophile response in accordance with the experience of others is considered a more dependable index of adrenal cortical stimulation.

The findings demonstrate that dihydroergocornine does not produce any significant inhibition of epinephrine stimulation of the adrenal cortex. That dihydroergocornine otherwise exerted adrenolytic and sympathicolytic activity in these experiments was indicated by reduction in blood pressure, modification of response to the cold pressor test and inhibition of epinephrine hyperglycemia. The combined use of dihydroergocornine with epinephrine or epinephrine-in-oil provides a method for therapeutic stimulation of the adrenal cortex, avoiding the undesirable side effects of epinephrine.

Autoantibodies in Different Phases of Human Glomerulonephritis

KURT LANGE, DAVID WEINER, MICHAEL M. A. GOLD, VICTOR TCHERTKOFF and VERA SIMON

The clinical facts which lead to the hypothesis that human glomerulonephritis is an organ specific antigen-antibody reaction will be reviewed together with the experimental evidence gained by heterologous antikidney serum injections. Using a further modification of Cannon and Marshall's collodion technique and its later modification by Cavelti, the authors have examined the sera of 51 nephritics of all stages and of 126 controls for the presence of antibodies to human kidney.

It was found that in 72.7 per cent of the tests done in cases of *all* stages of glomerulonephritis high positive titres can be obtained (average 1:412). The percentage of positive cases and the titres are higher in the later stages of glomerulonephritis than in the first month of the disease.

Twenty-eight determinations were done on 12 cases during the first month of the disease yielding an average titre of 1:107 with only 42 per cent of the cases being positive more than 60 per cent of the time.

Two hundred and twenty-one determinations were done in 39 cases after the first month of the disease yielding an average titre of 1:450 with 79.5 per cent of the cases being positive more than 60 per cent of the time. This difference may largely be due to the fact that during the first few days of acute glomerulonephritis no antibodies can be found in the serum since they are completely absorbed in the kidney.

Two hundred and five determinations performed on 126 normal controls yielded an average titre of 1:46 with only 17.5 per cent of the cases positive 50 per cent of the time or more.

Renal antigens obtained from infants or still-births show a much greater specificity and higher titres than antigens from adult renal tissue, a fact which may explain the high incidence of glomerulonephritis in young individuals.

The continuous presence of antibodies to kidney throughout all stages of the disease except the first few days forces one to assume that human nephritis does not result from a single insult with subsequent scarring but rather from the constant presence and occasional stimulation of antibodies to renal tissue.

Neutralizing these antikidney antibodies by means of kidney tissue emulsions or extracts should be carefully investigated as a possible therapeutic approach.

The use of antihistaminic drugs to mitigate the vascular effects of such an antigen-antibody reaction should also be investigated further.

A New Synthetic Anticoagulant (Heparinoid): Preliminary Report of Its Action in Humans

C. W. Sorenson, Joseph Seifter and I. S. Wright

Observations have been made of the effect in humans of a synthetic anticoagulant prepared by the sulfation of alginic acid. The material is a polysulfuric ester of polyanhydromannuronic acid obtained as a water soluble sodium salt which is stable at room temperature. Pharmacological data indicate that the drug has about the same toxicity in animals as heparin and is about one-tenth to one-twentieth as toxic as other synthetic polysaccharide sulfuric acid ester anticoagulants that have been tested. Rabbits given 5 to 15 mg./kg. of Heparinoid by intravenous injection every three hours daily for one week showed no toxic manifestations. Studies in animals on the fate of Heparinoid indicate that about twenty-five per cent is excreted in the urine. The remainder appears to be inactivated in the blood and the inactivated product picked up by the reticuloendothelial system.

The results being reported are based on the administration of Heparinoid to eleven patients, a total of twenty-five times. Observations have been concerned primarily with the anticoagulant action and toxic manifestations of the drug. It has been given intravenously in one, five and ten per cent solutions.

The anticoagulant effect has been measured by a modification of the Lee-White clotting time. The blood was drawn in siliconed syringes, placed in dry pyrex glass tubes, tipped gently at one minute intervals and the end point taken as the time when the blood held a vertical surface with the tube in a horizontal position. Duplicate tubes were run simultaneously and the clotting time reported was the average of the two tubes. Control times were nine and twelve minutes.

The amount of Heparinoid required to prolong the clotting time two to three times the control time was approximately 5 to 10 mg./kg. However, some individual variation in response was observed. This amount is approximately thirteen times that of the sodium salt of heparin necessary to produce comparable maximum clotting times. The duration of action of 5 to 10 mg./kg. given intravenously has been observed to last from 4 to 8 hours, but the results suggest that the action may be more prolonged than that of heparin given intravenously.

Immediate toxic reactions to the slow intravenous injection of Heparinoid have been observed in three patients: one of these was severe, two were mild. The severe reaction was manifested by nausea, vomiting, abdominal cramps, defecation, oppression over the chest, pallor, sweating, bradycardia, and fall in blood pressure to imperceptible levels. These symptoms and signs responded to prompt administration of epinephrine. This patient gave a history of similar but less severe reactions to intravenous papaverine and subsequently had a reaction to magnesium sulfate used in doing a circulation time.

One of the other reactions noted was a flushing of the face, feeling of epigastric fullness with belching, and a 16 mm. fall in the diastolic blood pressure. These manifestations disappeared without medication when the injection of the drug was discontinued.

The third reaction noted was a flushing of the face associated with a 10 mm. fall in blood pressure. The injection was completed and the flush subsided within fifteen minutes. Within about two hours both hands were noted to be slightly edematous; this lasted about two hours and subsided spontaneously.

No toxic effects of Heparinoid have been observed on the blood, kidneys, or liver as determined by red blood cell counts, white blood cell counts, sedimentation rates, urine examinations, P.S.P. tests, prothrombin times, B.S.P. tests, and cephalin flocculation, thymol turbidity, and total protein determinations.

Investigation is now under way on the clinical use of Heparinoid in the treatment of thrombo-embolic disease.

Fate of Dicumarol in Man: Relationship Between Prothrombin Time and Plasma Levels of Dicumarol

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The chief problem involved in dicumarol therapy is that of obtaining an adequate antithrombotic effect without hemorrhage. An understanding of the fate of dicumarol in the body might aid in the outlining of safer regimens of therapy.

A sensitive method for determining the concentration of Dicumarol in plasma and other biological fluids has been developed. Using this method, the absorption, excretion, distribution and rate of metabolism of dicumarol have been studied. The drug is almost completely transformed in the body, only traces being found in the urine after its administration. Its rate of transformation is slow with marked variability in different individuals. The drug is highly localized on plasma proteins and in various tissues, a fact which may explain its slow rate of transformation. Absorption of the drug from the gastrointestinal tract is slow and the rate varies with the individual and the dose.

Study of the solubility characteristics of the "apparent" dicumarol measured in the plasma, indicates that it is identical with authentic dicumarol. This finding, together with the extremely slow rate of transformation of the drug in vivo, makes it unlikely that its effect is directed through a derived product.

Prothrombin estimations were correlated with dicumarol levels. There is a threshold level for dicumarol below which no prothrombin time response is detected. Changes in prothrombin activity lag behind changes in dicumarol levels. However, the magnitude and duration of prothrombin time prolongation correlate much better with the dicumarol plasma levels than with the dose of dicumarol administered.

Some Limitations of Vagotomy in the Treatment of Peptic Ulcer: A Critical Follow-Up Analysis of Fifty Cases

MARTIN J. HEALY, JR. and PAUL K. SAUER

Fifty carefully selected cases of duodenal and marginal ulcer treated by vagotomy have been followed from six months to two and one-half years, and results evaluated. All fifty received transthoracic vagotomy without other surgical measures, with one postoperative death. Each case met the following criteria: intractable ulcer pain, failure of medical treatment, X-ray evidence of active ulcer, and absence of gastric retention. According to insulin (Hollander) tests, vagotomy was complete in 82 per cent, incomplete in 8 per cent, equivocal in 4 per cent and undetermined in 6 per cent. Early failure occurred in 10 per cent of the cases as a result of unrelenting gastric atony which necessitated further surgery within three and one-half months after vagotomy.

Careful follow-up studies were made in all cases. Ninety per cent of the cases were readmitted to the hospital at intervals for clinical evaluation, secretion studies and X-rays. Information regarding the remaining 10 per cent was secured from other hospitals or by questionnaire.

After an average follow-up period of 18 months, interim evaluation of results is as follows:

Effect on Ulcer Symptoms

Excellent (Symptom free)	18%
Good (Symptoms improved)	36%
Fair (Temporary or incomplete relief)	12%
Failure	34%
Early from atony	10%
Late, full recurrence	24%

Nine (18%) of the failures required further surgery from three weeks to twenty-seven months after vagotomy. Seven had gastric resection and two had gastroenterostomy performed. Three of these cases subsequently developed marginal ulcers despite apparently complete vagotomy.

Effect on Ulcer as Demonstrated by X-ray

Active Ulcer—	
Niche and/or deformity, irritability, spasm, and tenderness	38%
Inactive ulcer—	
Deformity alone	42%
Early failure	
No X-rays after three months	8%
Unknown	12%

Secondary prolonged disturbances in gastric motility were observed on X-ray examination as follows:

Unremitting atony requiring early surgical intervention	10%
Prolonged atony	
>100% enlargement	
>50%—6 hr. retention	6%
Moderate atony	
>30% <100% enlargement	
<50%—6 hr. retention	30%
Slight atony	
<30% enlargement	
No retention	6%
Normal stomach	
No change observed	36%
Unknown	12%

According to the authors' data, a patient who is subjected to vagotomy has one chance in five of securing complete relief, two chances in three of being benefited to some extent, and one chance in three of failure, all within an average of eighteen months postoperatively. For this rather dubious benefit he must accept a small but nevertheless significant mortality risk and the unhappy prospect that his continued freedom from ulcer symptoms is by no means assured. Half the time he must also pay an additional premium in the form of persistent symptoms of gastric atony which not infrequently may be more disabling than the original ulcer.

On the whole, the authors feel that vagotomy, even in carefully selected cases, has not proven successful in a sufficient number of cases to justify its routine use. Despite the lower operative mortality, failures are too frequent and morbidity too serious after vagotomy to justify its replacement of other more effective methods of surgical treatment.

A Method for the Preparation of Exteriorized Skin Covered Loops of Intestine for the Study of Bowel Obstruction

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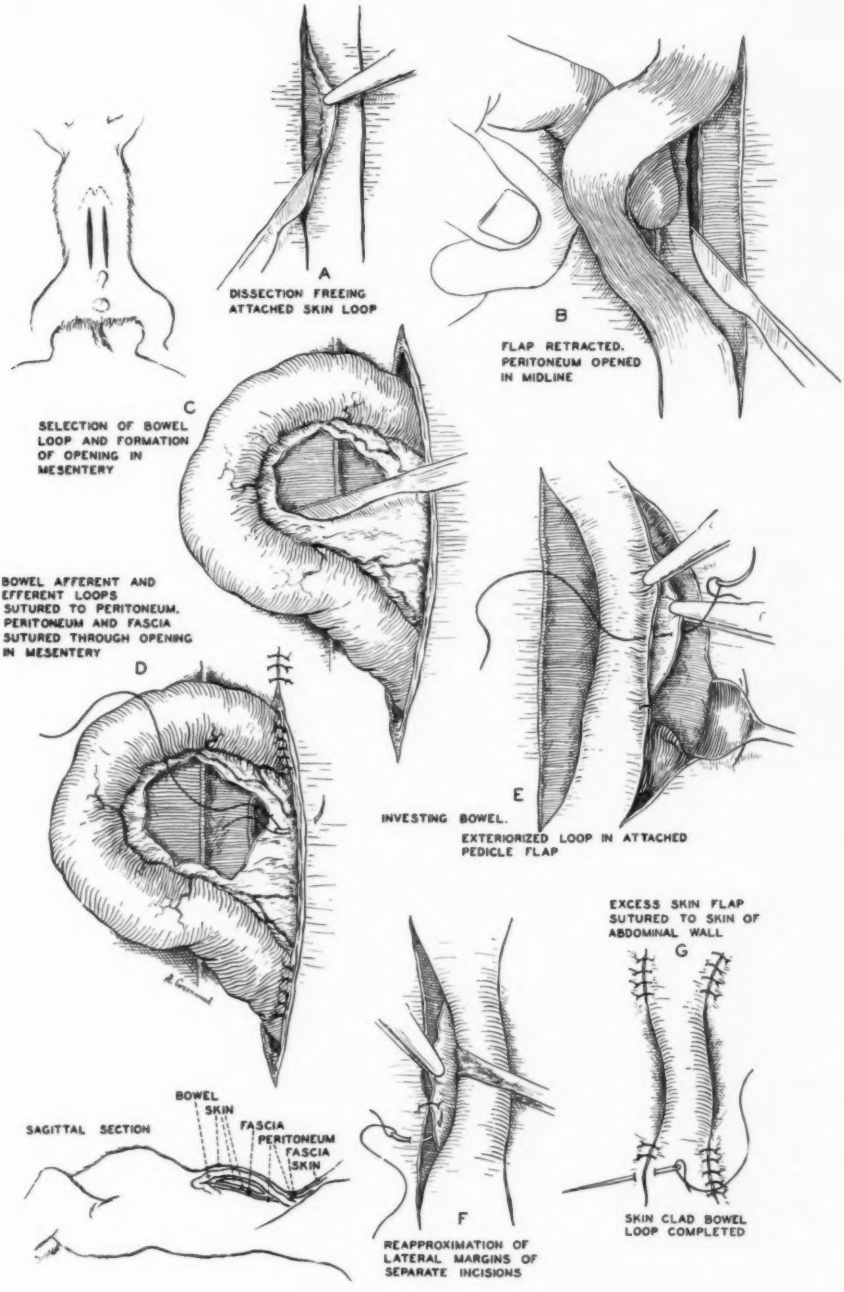
In the course of certain experiments concerned with intestinal obstruction it is advantageous to prepare exteriorized loops of intestine which can be obstructed for varying periods of time and then released without the necessity of repeated operations. The following preparation can serve such purpose in that the animal can be kept in a normal state for long periods of time and may be used for study at will. There is no exposure of naked loops of intestine and the continuity of the gut or its nerve or blood supply is not interfered with.

Two straight incisions placed 2½ inches


A
DISSECTION FREEING ATTACHED SKIN LOOP
B
FLAP RETRACTED. PERITONEUM OPENED IN MIDLINE
C
SELECTION OF BOWEL LOOP AND FORMATION OF OPENING IN MESENTERY
BOWEL AFFERENT AND EFFERENT LOOPS SUTURED TO PERITONEUM. PERITONEUM AND FASCIA SUTURED THROUGH OPENING IN MESENTERY
D
E
INVESTING BOWEL.
EXTERIORIZED LOOP IN ATTACHED PEDICLE FLAP
SAGITTAL SECTION
BOWEL
SKIN
FASCIA
PERITONEUM
FASCIA
SKIN
F
REAPPROXIMATION OF LATERAL MARGINS OF SEPARATE INCISIONS
EXCESS SKIN FLAP SUTURED TO SKIN OF ABDOMINAL WALL
G
SKIN CLAD BOWEL LOOP COMPLETED

on either side of the midline are made. These incisions extend down through the skin and subcutaneous fascia, permitting a layer of skin to be raised which is attached at the upper and lower ends. The abdomen is opened through a midline incision beneath the segment of skin. The loop of bowel to be exteriorized is then brought out and the peritoneum, muscle, and fascia closed about the loop of intestine through its mesentery. It is necessary to suture the peritoneum through an opening in the mesentery made in the area where there are no vessels or nerves. The approximation of the peritoneum, muscle, and fascia must be brought loosely about the vessels so as not to cause constriction. The flap of skin which has been raised from the midline as a pedicle graft is then approximated about the loop through the aperture in the mesentery. A free space is left which traverses the mesentery and the skin in this area so as to permit the application of a clamp or other means of obstructing the bowel. The skin on either side of the midline incision is then undercut sufficiently to allow approximation to the edges of the skin which also pass through the aperture in the mesentery together with the peritoneum and fascia.

It can readily be seen from the diagram which is presented (see figure) that this preparation permits of sampling from the intestine by the insertion of a needle through the skin and gut which lies beneath it. If such sampling is carried out, the needle ought to be extended tangentially for a distance through the skin and the loop in order that intraenteric pressure will tend to close the needle hole without leakage.

When it is decided to produce a simple obstruction, all that is necessary is to apply a clamp at one end of the loop. If a closed loop obstruction is desired, a clamp may be placed on both ends of the loop.

Studies to determine the relation of the autonomic nervous system to intestinal obstruction are being undertaken. This preparation lends itself readily to experiments and because of its simplicity it may be of considerable use to others who are carrying on similar studies.

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*The Biological Chemistry of Wound Healing: Effect of Blocking the Sulfhydryl Group (-SH) in Vivo with Iodoacetic Acid**

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Previous studies have shown that protein depletion impedes the rate of healing of laparotomy and of surface wounds in rats. It has also been shown that dl-methionine administered parenterally following infliction of the wound serves to return the curve of wound healing toward normal. The cause and nature of this effect of methionine has been sought. Possible hypotheses include the following: 1) Liver function may be improved by the lipotropic effect of the methionine. 2) The healing wound may require the specific amino acid methionine as a raw material in a quantity beyond that available to it by catabolism of its own tissues in the protein-depleted state. 3) Since the skin and connective-tissue are relatively rich in organic sulfur, the sulfhydryl portion of the methionine molecule may be essential to the normally healing wound.

The last hypothesis has been tested by chemically blocking the —SH radical with parenterally administered iodoacetic acid. This compound has been shown to inhibit the sulfhydryl group of various enzymes and other compounds in vitro and in vivo. It also inhibits growth in normal young rats.

A series of albino rats received iodoacetic acid for a total of 10 days immediately pre-

ceding and following infliction of a standard laparotomy wound. Groups of animals were killed on the 3rd, 4th, 5th, and 6th days postoperatively, and the tensile strength of the wounds was tested. A significant lowering of the tensile strength was noted when compared with normal rats.

A third group of rats was tested which were in every way identical with the first group except that in addition to iodoacetic acid they were given an excess of dl-methionine, a physiological precursor of sulfhydryl-containing compounds. These rats showed acceleration of healing when compared with the iodoacetic acid-treated rats.

It is concluded that blocking the —SH radical in vivo impedes wound healing in otherwise normal rats. The possible site of this blocking effect is briefly discussed as is its significance to the wound-healing problem.

Starch Sponge—A Hemostatic Agent

SAMUEL S. ROSENFELD

A number of highly efficient blood coagulants have recently been introduced into medical practice and have been found of great value. The reasons that impelled the author to describe yet another hemostatic agent is its demonstrated effectiveness and low cost.

In 1947 a patent was granted the U. S. Department of Agriculture for the manufacture of an absorbable starch sponge. The dry sponge is capable of absorbing sixteen times its weight of water, and the department was interested in it because it could absorb appreciable amounts of medicaments such as penicillin, gramicidin, sulfapyridine, etc. and is slowly absorbed from body cavities.

It seemed to the author that the chemical and physical nature of the starch sponge made it highly probable that it also possessed blood coagulating properties.

Numerous experiments have demonstrated that the starch is taken up by the peritoneum and is metabolized like any ingested starch. No sensitizing or anaphylactogenic properties were encountered. The majority of investigators state that the tissue reaction to starch is not significant. Our own experiments confirm this statement. We left large pieces of dry and wet sponge amongst coils of the intestine without opening any large vessel. At autopsy gross inspection in the majority of these animals revealed none or negligible adhesions.

Taking into consideration the fact that the vena cava is located in the retroperitoneal space, the cellular tissue of which is almost embryonal in its reaction to stimuli, the adhesions we found at autopsy (usually three to four weeks after operation) were surprisingly moderate.

The blood coagulating properties were demonstrated by us on rabbits, using suitable controls. We employed the large veins of the ear, the femoral vein, and the inferior vena cava.

In every case where the starch was properly applied to any of the above-mentioned incised vessels, including the vena cava, the animal survived. All controls died when the larger vessels were opened.

To date, we have used the starch clinically 76 times to control bleeding. In 35 instances, the bleeding was intraperitoneal; in 29 cases, it was vaginal; in 10 patients cervical and in 2 rectal. The procedures in which these applications were made included: abdominal and vaginal hysterectomy, myomectomy, ruptured corpus luteum cyst, and cesarean section. In some of the cesarean cases the starch was introduced into the uterine cavity through the uterine incision.

The starch added greatly to the safety of the operation, especially in hysterectomy where the oozing was easily controlled, thus obviating clamping and possible ureteral injury.

There was no mortality in this series.

There was no morbidity that could actually be attributed to the starch.

Starch sponge is an efficient hemostatic agent. Its effects appear to be purely local and its hemostatic action is probably due to its gel formation, which permits it to absorb many times its weight in blood. This property causes the sponge to swell and exert pressure on the bleeding points. An additional barrier to blood loss is its adhesive quality.

An important factor is its low cost. It can be produced for as little as 7 cents a pound.

Management of Ascites Due to Cirrhosis of the Liver; the Use of "Rice" Diet, Blood and Plasma, Diuretics and Surgery

GEORGE F. KAMEN, MAX TRUBEK and
JERE W. LORD, JR.

From New York University Post-Graduate Medical School and Fourth Medical and Surgical Divisions of Bellevue Hospital

Kempner has demonstrated that hypertensive patients maintain protein balance on the rice-fruit-sugar diet which contains approximately 470 gms. of carbohydrate, 20 gms. of protein and 5 gms. of fat per 2,000 calories. Further, this diet is low in salt, having less than 150 mgms. of sodium and 200 mgms. of chloride.

It occurred to us that the cirrhotic patient with ascites might obtain benefit from the "rice" diet by virtue of its low salt composition, particularly when a protein hydrolysate low in sodium and high in tryptophane (120 gms. of powder contain the equivalent of 70 gms. of protein) was added daily. The sodium content of the powder is less than 50 mgms. per 100 gms. In addition, anemia and hypoproteinemia were treated with transfusions of whole blood or plasma and daily vitamin supplements were supplied. In several patients diuresis was achieved only by the use of intramuscular mercuhydrin given concurrently with blood or plasma infusion.

Six patients suffering from intractable ascites due to cirrhosis of the liver have been managed by the "rice" diet and the early results to date have been encouraging. One patient, a 36 year old white man, entered the Fourth Medical Division of Bellevue Hospital on October 4, 1948, complaining of ascites of 2 months duration, abdominal pain, anorexia and weakness. A diagnosis of cirrhosis of the liver of alcoholic-nutritional etiology was established and the usual high protein high carbohydrate, low fat, low salt diet was instituted. Anorexia, ascites requiring paracentesis, and weakness persisted. Of poor prognostic significance from past experience, were the anemia, a large smooth liver and the febrile course. The patient's course was slowly downhill, in spite of the use of diuretics and the "Patek regime" until November 18, 1948 when the rice-fruit-sugar diet, with the addition of the oral protein supplement was instituted. During the next 7 weeks the patient lost 33 pounds in weight. his abdominal circumference decreased from 100 cms. to 72 cms. and his appetite and strength improved. Prior to the institution of the "rice" diet, the patient had had a massive gastrointestinal hemorrhage requiring 3 transfusions. A gastrointestinal roentgen series failed to reveal esophageal varices or a peptic ulcer. The patient has been free from ascites for 3 months and in good condition with the exception of a small residual left hydrothorax which shows no evidence of a neoplastic or tuberculous etiology. A high red count and hemoglobin and normal blood protein fractions have been sustained.

Of the remaining 5 patients, one was showing marked improvement on the regime but succumbed to a massive gastrointestinal

hemorrhage during the second month of therapy. Three others have not required paracenteses while on the diet over periods of 4, 3 and 2 months respectively. The sixth patient is a 43 year old man with portal cirrhosis on an alcoholic nutritional basis who required approximately 40 taps over a period of 10 months. During one month of the regime no paracenteses were required, although some ascites persisted. Because an esophagram showed esophageal varices, the patient was subjected to a portacaval shunt (an end-to-end anastomosis of the portal vein with inferior vena cava, through a right sided thoraco-abdominal approach) with a lowering of the portal pressure of 475 to 190 mms. of water when the shunt was opened. His post-operative course was smooth except for 3 or 4 days of intermittent mental confusion.

In summary, the "rice" diet supplemented by certain important factors such as added oral salt-free protein, vitamins, concurrent use of plasma and blood transfusions with mercurial diuretics proved to be an effective form of therapy in the management of intractable ascites due to cirrhosis of the liver. In one instance the above regime proved to be excellent for the pre-operative preparation of a patient subjected to a portacaval shunt.

Carbohydrate Utilization in Surgical Patients: The Blood Lactic Acid in Pre- and Post-Operative Patients After the Administration of Glucose

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Abridgement of thesis submitted to the Faculty of the Graduate School of the New York Medical College in partial fulfillment of the requirement for the degree of Master of Medical Science in Surgery

During the post-operative period glucose is the chief source of nutrition. It is, therefore, essential to determine whether the glucose given is actually utilized or simply stored. Many studies on this subject indicate that the coefficient of correlation for grams of glucose given and grams retained is high. Unless the sugar is burned, however, post-operative starvation ketosis and hypoproteinemia will occur.

That glucose is capable of immediate oxidation following absorption is a general assumption. Koster, however, in 1930 used the respiratory quotient as the index of carbohydrate combustion and found that a normal rise in the respiratory quotient following the intravenous injection of glucose did not occur until the fifth post-operative day. He concluded that a carbohydrate metabolic disturbance existed during this period. His work has never been duplicated or challenged.

The psychic and organic effects of major surgery can cause marked alterations in the bodily metabolic processes. The classical demonstrations of Cannon indicate the psychic effects upon the adrenals and Seyle and others have shown the influence of trauma, blood loss and shock on the carbohydrate metabolizing hormones.

The use of the respiratory quotient as a criterion of carbohydrate utilization has been violently attacked by Soskin and Levine. The use of the blood lactic acid response to glucose as an indicator has gained general recognition. Root and Carpenter in 1946 checked their previous observations concerning the ability of the diabetic to burn sugar using the blood lactic acid.

In order, therefore, to determine whether the post-operative period is characterized by inadequate sugar metabolism, it was decided to utilize the lactic acid of the blood as an indicator.

Method: Ten patients were studied in the pre- and post-operative periods. Pre-operative results were used as the norm. All patients were given high doses of vitamins

daily before study. Patients who were febrile or who showed kidney or liver damage were excluded. Young and old patients of both sexes in good, fair and poor general condition were included. The operative procedures varied widely in type and duration. The amount of each anesthetic agent administered was estimated.

A 300 cc. aqueous solution containing 50 gms. of glucose was given intravenously during an interval of twenty minutes. All patients were in the post-absorptive state. The blood lactic acid and blood sugar as well as the cephalin flocculation and thymol turbidity were determined from a sample of blood withdrawn just before the infusion of glucose. These served as the base-line values. Blood was withdrawn every thirty minutes for two hours thereafter for lactic acid and sugar determinations. The total amount of sugar excreted in the urine was determined quantitatively. The fluid intake and output were carefully recorded.

Results: There was an increase in the blood lactic acid in all patients in both the pre- and post-operative periods following the intravenous administration of glucose. The average rise noted during both periods was of approximately the same magnitude. The rise also corresponds with that observed in normal subjects by other investigators. Experiments were carried out for from two to 17 hours post-operatively. The maximum rise in lactic acid did not occur regularly within a constant period after glucose administration. This confirms the observations of others that the lactic acid cannot be correlated quantitatively with the blood sugar curve. The rise does indicate, however, that glucose was definitely in the process of oxidation in all patients during the post-operative period.

Blood sugar levels post-operatively were slightly higher than the pre-operative values. Approximately three times as much sugar was lost in the urine of patients post-operatively. Fluid balance data did not show that hypertonic glucose solutions cause excessive fluid loss post-operatively. This observation has also been made by other investigators.

CONCLUSIONS

1. Lactic acid, an intermediary compound in carbohydrate metabolism, has been studied in pre- and post-operative patients following the intravenous administration of glucose.

2. The blood lactic acid increase observed in all patients demonstrates that glucose is capable of oxidation and utilization in the pre- and post-operative periods.

3. Glucose was not found to be associated with excessive glycosuria or fluid imbalance when administered intravenously in the immediate post-operative state.

Studies with the Quantitative Cephalin-Cholesterol Flocculation Reaction

Effect of variation in temperature at which Hanger reaction occurs. The protein patterns of normal and abnormal liver-disease sera

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In a previous publication (J. Clin. Invest. *27*, 737 (1948)) a method was described for determining small variations in the cephalin-cholesterol flocculation reaction (Hanger) by means of *quantitative* cholesterol determinations. The results are expressed as cholesterol units. By means of this procedure the effect of temperature on a large number of normal and abnormal sera were studied. A marked variation was found in the amount of flocculation obtained with the same serum at different temperatures.

These temperature studies indicate that all human sera with suspected liver disease

can be divided into four serum protein patterns based on the cholesterol unit values obtained at 4°, 25°, 37.5° and 56° C. These patterns are classified on the basis of severity of liver damage as follows:—

Type I: Normal sera
Type II: Slight liver damage e.g. chronic hepatitis
Type III: Moderate liver damage e.g. acute hepatitis
Type IV: Severe liver damage e.g. cirrhosis

Serial studies on a single case of acute infectious hepatitis from the peak of the attack to date of discharge, show a gradual change in the pattern from Type IV to Type I as the clinical condition of the patient improved.

Cases of infectious mononucleosis or malaria, which are often associated with liver damage, exhibit the same serum protein patterns as do cases of infectious hepatitis. Similar liver disease patterns were found in some cases of multiple sclerosis.

These changes in the serum protein patterns obtained with this simple technique in various liver disease cases parallel those found by other investigators employing the more elaborate electrophoretic procedures.

Hemophilioid Disease

A hemorrhagic disease with prolonged coagulation time and a circulating anticoagulant. Report of a case in a female

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In 1940 Lozner, Jolliffe and Taylor were the first to recognize a hemophilia-like disease associated with a circulating anticoagulant. There have been a total of ten reported cases; of these, two were in women. The present study is concerned with a young woman who manifests this disease.

F. L., a thirty year old white female with no past or familial bleeding history, had an uneventful second delivery in November 1947. Seven weeks later she had a mild siege of pains in the thigh and the popliteal region, slight fever, and leukocytosis. Three months post-partum she developed a large hematoma in her right jaw following a dental procedure. In March 1948 ecchymoses and pain in the left leg appeared. This was accompanied by a prolonged coagulation time of 39½ minutes (normal 6-12) of the venous blood. The blood picture was otherwise unremarkable; likewise the platelets, prothrombin, tourniquet test, bleeding time, clot retraction, calcium, and fibrinogen were all normal. In July 1948 renal colic and hematuria occurred. The hematuria did not respond to transfusions, so that the patient had to be hospitalized during August 1948. Anti-hemophilic globulin, protamine, and fresh blood serum, as well as whole fresh blood, were administered but these had no effect upon the hematuria or the prolonged clotting of the blood.

Special studies were made to determine the underlying cause of the abnormal clotting:

1. The patient's blood was mixed with normal blood and was found to prolong the coagulation of the normal blood. This indicated the presence of anticoagulant in her blood.
2. The anticoagulant factor was present in plasma and serum as well as in whole blood.
3. Ammonium sulfate fractionation of the plasma indicated its presence in the pseudo-globulin fraction.
4. It was thermostable at 56° C.
5. It was not dialyzable.
6. The electrophoretic curve of the plasma was normal.
7. The anticoagulant was not neutralized by protamine, toluidine blue, or anti-

hemophilic globulin.

8. There was no increase of anti-thrombin.

9. Conversion of prothrombin was not delayed.

10. Factor VI of Owren was normally present.

11. Titrations against rabbit and human thromboplastin showed a deficiency of thromboplastin in the patient's plasma.

12. The anticoagulant was directly antagonistic to anti-hemophilic globulin, completely neutralizing the effect of the latter upon known hemophilic blood.

13. Additional studies indicated that the anticoagulant might be an antibody to some factor (such as anti-hemophilic globulin) in normal plasma, but this could not be confirmed by precipitin and complement-fixation tests.

After discharge from the hospital, the patient improved gradually and felt better. She was last seen on February 11, 1949, thirteen months after the onset of her illness. The hematuria had subsided, but there were still occasional slight ecchymoses. Her coagulation time was still prolonged (76 minutes) and the anticoagulant was still present.

Aureomycin and Chloromycetin in Brucellosis

With special reference to chronic brucellosis

HAROLD J. HARRIS

The diagnosis of low-grade chronic *Brucella* infection often lacks cultural proof. Evaluation of treatment methods is difficult. Any effective therapy which is free of deleterious effects and which obviates the need for hospital care is worthy of investigation.

AUREOMYCIN

Aureomycin hydrochloride was given orally to 110 patients, all but one with culturally-negative chronic brucellosis.

Dosage was 3.0 to 4.0 grams daily in the earlier cases and 2.0 grams daily in later cases, in 4 equally spaced amounts. Total dosage was from 18 to 25 grams in 7 to 14 days, if tolerated. Dosage in children was proportionately lower.

Side effects of a frequency, kind and severity not previously reported occurred in 48 of the 79 females (60.7 per cent) and in 6 of the 31 males (19.3 per cent), a notable difference between the sexes.

Headache, nausea, vomiting, epigastric pain, persistent diarrhea and anal irritation (with fissuring in 2) occurred in either sex. Mucous membrane manifestations of the mouth, throat and pharynx with perleche-like lesions, occurred in 12 females and in 1 male, within 3 days in some. Vaginitis occurred in all of the 12 females, with slight bleeding in 3. Bladder irritation occurred in 8, fleeting urticaria in 3, dermatitis of the hands in 2 and slight generalized desquamation of the epidermis in 2. Peculiarly voracious and capricious appetites, especially for protein foods, were noted in many.

Thirty-four patients were unable to tolerate aureomycin for more than a few days but six were able to take smaller dosage of a more highly refined product.

The high acidity was presumed to account for some of the gastrointestinal and bladder manifestations, and destruction of the normal intestinal flora and/or allergy for the mucosal and dermal reactions.

Previously existing symptoms were temporarily aggravated in many patients.

Deleterious effects definitely ascribable to aureomycin were not noted. Agranulocytosis developed on the sixth day in a child who had been moribund at initiation of treatment; however, there had been a steadily developing leucopenia and neutropenia prior to treatment; recovery followed. Perforation of an unsuspected pre-pyloric ulcer, pos-

sibly due to direct irritation, occurred in one patient, with spontaneous healing.

Results of treatment cannot be fully evaluated until after prolonged observation. Of 55 patients who had adequate dosage and were observed for from 1 to 6 months following treatment 49 (89 per cent) made from partial to complete recovery; 23 required two or more courses. Six patients (11 per cent) showed no improvement.

In the one culturally proved case, there was rapid recovery but cultures were positive after each of two courses, with minimal evidence of relapse; following a third course cultures have remained negative for 3 months but minimal symptoms probably referable to Brucella infection continue to recur.

CHLOROMYCETIN

Chloromycetin was given to 25 patients (20 females and 5 males), all culturally-negative, in dosage of 2 to 4 grams daily with total dosage of 25 grams.

Side effects of appreciable degree occurred only in female patients previously intolerant of aureomycin; 5 of these had recurrence of mucous membrane involvement and 2 had to discontinue it because of gastroenteritis. Relative freedom from side effects allowed use of chloromycetin in 13 patients who could not take adequate amounts of aureomycin. Fleeting sunburn-like erythema occurred in 3 patients, urinary frequency in 3.

Results of treatment were apparently comparable to those from aureomycin. Of the 14 patients adequately treated and observed, 11 (78.5 per cent) made from partial to complete recovery during observation periods of from 1 to 4 months; 3 (21.5 per-cent) showed no improvement. Repeated courses were needed in 3 patients.

CONCLUSIONS

Aureomycin and chloromycetin favorably influence the course of brucellosis. It is premature to discuss curative effect. Chloromycetin is attended by fewer and less severe side effects. Male patients show greater tolerance. Effect on the intestinal flora is profound. Optimum dosage remains to be determined.

Response of Gastric-Dissolved Mucoprotein to Insulin: A New Test for Evaluation of Secretory Status of Fundal Glands and Integrity of Nervous Pathways to the Stomach

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In previous studies the authors showed that the dissolved mucin of the gastric juice was a mixture of at least two chemically different compounds, which they separated and called "dissolved gastric mucoproteose" and "dissolved gastric mucoprotein."[1] A chemical colorimetric method for quantitative determination of both fractions was developed,[2] and evidence was obtained to show that the mucoproteose was mainly a soluble split product of the enzymatic digestion of the visible surface epithelium mucus, while the mucoprotein fraction was the secretory product of mucous cells of glands of the fundus and corpus of the stomach.[1]

The authors showed also that the secretion of mucoprotein fraction was strongly influenced by vagal impulses and that humoral stimuli effective for acid secretion were almost devoid of any effect on secretion of gastric mucoprotein.[2] Insulin, injected intravenously in the dose of 16-20 units is a powerful stimulus for secretion of mucoprotein and after 40-60 minutes causes a sharp rise in mucoprotein concentration, the output exceeding the fasting

level 3 and more times. This rise coincides with the peak of hypoglycemic symptoms (drowsiness, dizziness, perspiration, fall in blood sugar) and is abolished if the nervous pathways to the stomach are divided or by atrophy or destruction of the mucous cells of gastric glands of the fundus and corpus area.

Three main patterns were observed in regard to mucoprotein and acidity response to insulin in over 50 tests performed on 40 individuals: 1) Positive pattern, characterized by a definite rise in mucoprotein (more than 60 per cent) and acidity; this is typical for normal stomach, nervous indigestion and gastro-duodenal ulcer. 2) Dissociated pattern, characterized by positive mucoprotein but negative or low acid response: this is found in subtotally resected stomach and in localized antral lesions, which impair the secretion of acid but leave intact the site of mucoprotein secretion, i.e. fundal area. 3) Negative pattern, characterized by a low or abolished response of mucoprotein and acidity to insulin; this is found in diffuse atrophic lesions of the gastric mucosa, in diffuse destruction of gastric glands by invading carcinoma, and as a rule after complete vagotomy.

Since the response of mucoprotein to insulin is abolished by vagotomy but not by subtotal gastrectomy, the mucoprotein test is also suggested for evaluating the completeness of vagotomy, especially in cases in which vagotomy is combined with subtotal gastric resection, and when it is impossible to decide whether a negative Hollander test depends upon vagotomy or resection itself.

Insulin test: The stomach under fasting conditions is emptied with Levine tube as completely as possible and 20 minutes later the second fasting specimen is again aspirated completely. Insulin is injected intravenously in a dose of 16 units (in thin and young patients—12 units only) and the stomach is emptied as completely as possible 20, 40 and 60 minutes after injection. Each of 5 specimens is tested for acidity and mucoprotein concentration. The mucoprotein and acidity curves are drawn on the basis of these determinations.

Volumetric office procedure for quantitation of gastric mucoprotein. To 4 cc. of centrifuged gastric juice 2 cc. of 10 per cent trichloracetic acid is added; after standing for a short time the mixture is centrifuged and the supernatant fluid is separated. Into a 15 cc. centrifuge tube graduated in 0.05 cc. or 0.1 cc. markings, 5 cc. of supernatant fluid and 7½ cc. of acetone are placed. The content of the tube is mixed and the tube is set in the water bath or incubator at 56° C. for ½ hour or at 40° C. for 1 hour. Then the tube is centrifuged for about 10 minutes, the supernatant fluid is briskly decanted and the remaining mucin precipitate is dissolved in 2 cc. of 1/10 N sodium hydroxide, using the wooden applicator for stirring. To this 3 cc. 1/10 N hydrochloric acid and 5 cc. distilled water are added, the content is mixed, left for 20 minutes at room temperature, and centrifuged exactly at 3000 RPM for 5 minutes (stopwatch). The volume of the precipitate is read on the graduations using magnifying glass, and the mucoprotein concentration is read from the table (interpolating intermediate values if necessary), as follows:

Calculation Table

Volume of precipitate in cc. after 5 min. of centrifugation at 3000 RPM.	*Concentration of gastric mucoprotein in mgm. per 100 cc. fluid gastric juice*
0.05	23
0.10	50
0.15	85
0.20	115
0.25	150
0.30	185
0.35	220
0.40	260
0.45	300
0.50	350
0.55	400
0.60	450

This simplified volumetric quantitation of mucoprotein was standardized on the basis of the original colorimetric tyrosine method published for quantitation of mucoprotein and mucoproteose fractions.[2]

CONCLUSIONS

Because the secretion of gastric mucoprotein is mainly dependent upon vagal influences and is almost independent of humoral stimuli, and because it depends on the status of effector organ, in this case the gastric glands of the fundus and upper corpus of the stomach, the testing of the mucoprotein response to insulin supplies the information which cannot be obtained by means of other available tests, and is suggested as a new functional test of the stomach.

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Electrolyte Abnormalities in Chronic Congestive Heart Failure; Effects of Administration of Potassium and Sodium Salts

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Plasma, edema fluid and urine electrolytes were studied during congestive failure. Plasma sodium was significantly below normal in twenty-seven of thirty patients. Plasma chlorides and potassiums ranged from below to above normal; chlorides, however, were relatively increased. Urinary sodium was low whereas potassium was high. Repeat measurements after recovery in five patients showed that the low sodiums had increased whereas chloride and potassium shifted toward normal.

During mercurial diuresis sodium chloride excretion increased but sodium in urine was usually hypotonic; chlorides, however, equalled or exceeded sodium. Potassium excretion increased fivefold.

The data indicated that failure is frequently associated with hypotonic extracellular fluid excessive in chloride and that during recovery, relatively more water, chloride and potassium are excreted than sodium. The possibility that some of the water excreted was of intracellular origin is also supported by the rise in the noncolloidal extracellular osmotic pressure which might draw water from cells.

Accordingly, sodium lactate was given to raise extracellular sodium and bicarbonate concentrations, and potassium acetate to raise intracellular potassium and bicarbonate. Up to 400 mEq of each were administered orally for periods up to thirty days to six patients in failure. Mercurial diuretics previously required were omitted. Loss or no gain in weight occurred despite positive balances of sodium and potassium. Sodium excretion increased markedly although subnormal plasma sodium initially decreased further. Shift of water from intra- to extracellular compartments is thereby suggested.

Individual administration of the chloride and acetate of Na or K increased weight two to six kilograms; therefore mercurials were required after each single salt period. Simultaneous administration of sodium chloride and potassium acetate caused minimal weight gain despite positive Na, Cl, and K balances.

Extracellular hypotonicity may be associated with intracellular hypotonicity and depletion, hence separate readjustment of either compartment augments edema. Intracellular abnormalities may also be important in failure. Appropriate electrolyte administration may be of value in controlling edema in congestive heart disease.

*The Role of Proteins in the Pathogenesis of Renal Insufficiency**

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Reports from several sources have indicated that high molecular weight proteins (hemoglobin) and low molecular weight proteins (myoglobin, Bence-Jones protein) are capable of producing fatal renal insufficiency. Studies with hemoglobin and myoglobin have already established these substances as potent agents in the production of acute renal necrosis. A recent report incriminates the former substance as the cause of chronic renal insufficiency resulting from repeated renal insults, suggesting similar mechanisms in myoglobinuria. Intermittent injections with Bence-Jones protein in relatively small amounts has not resulted in renal impairment. No studies with large amounts of globulin have been carried out in either humans or animals. Accordingly, rabbits were injected with homologous (rabbit) gamma globulin procured through the courtesy of the Lederle Laboratories as Rabbit Anti-Pneumococcus Serum (Refined) which proved to be a 10 per cent solution of almost pure gamma globulin.

The experiences with rabbits injected intravenously and intraperitoneally is presented. The quantity of urine voided in 24 hours was measured for globulin content. Concomitant blood counts and hematocrit readings were recorded in several instances. Histologic studies of the kidneys performed post-mortem revealed evidence of intrarenal hemorrhage due to the Silverman needle biopsy technique.

Following either intravenous or intraperitoneal injections of gamma globulin significant changes in the serum protein levels were observed. The details of individual experiments indicate a pronounced fall in the serum albumin as the globulin level rose (usually after 4 or 5 daily intraperitoneal injections of 50 cc. each). In one animal which received prolonged and repeated intraperitoneal injections, glomerular changes were observed.

* Work done under a grant from the Joseph and Helen Yeamans Levy Foundation in memory of Miriam Levy Finn.

The Use of Urinary Pigment Excretion for the Measurement of Basal Metabolic Rate

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The diagnosis and control of hyperthyroidism depends, to a large degree, upon an accurate determination of the basal metabolic rate. It is known that such conditions as neurocirculatory asthenia and apprehension may cause an increase in oxygen consumption which does not reflect the true basal metabolism. It is virtually impossible to obtain accurate BMR determinations in patients without teeth or with perforated ear drums. The existence of these conditions which produce an increase in oxygen consumption rate, in the absence of an increased basal metabolism, frequently renders the accuracy of the BMR, as determined by the respiratory calorimeter, open to question. Since the determination of the true basal metabolism, and not a machine-measured increase in oxygen consumption, is of great importance in the diagnosis and control of thyrotoxicosis, the presence of any of the aforementioned conditions fre-

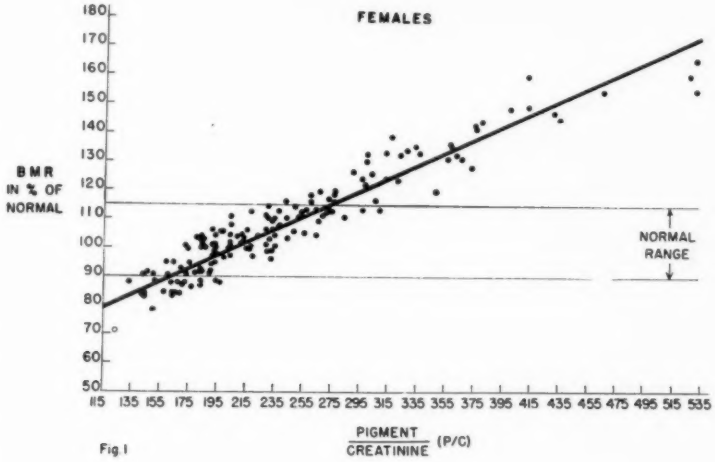

Fig. 1

quently makes it impossible to determine true basal metabolism, with our present methods. The elaboration of a simple clinically-applicable procedure, which would serve as a measure of basal metabolism, independent of oxygen consumption at the time of the test, is, therefore, of great importance.

Drabkin[1] has shown that the output of the urinary pigment, urochrome, is remarkably constant from day to day, is independent of diet, but bears a relation to the level of basal oxygen consumption, and is therefore, a product of endogenous metabolism, being increased in hyperthyroidism and diminished after extirpation of the thyroid.

The daily creatinine excretion is extraordinarily constant for the individual, it is not influenced by ordinary exercise, and is practically independent of the protein level of the diet.

Ostow and Philo[2] reinvestigated the nature of the relationship between urinary pigment output and the basal metabolic rate. To correct for weight, height, and habitus of the subjects, the rate of pigment excretion was compared to the rate of creatinine excretion. This ratio (P/C) was found to parallel the basal metabolic rates

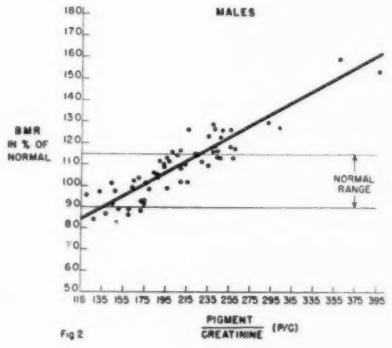

Fig. 2

of patients to whom thyroid extract was administered.

The purpose of this investigation was to determine whether an accurate correlation existed between basal metabolic rates as determined by the respiratory calorimeter and the P/C ratio, and, if possible, to obtain a regression equation from this relationship. If a statistically valid equation could be derived, the P/C ratio could be applied wherever the accuracy of the BMR was in question.

The urinary pigment and creatinine concentrations are determined with a spectrophotometer.

Results obtained from 156 adult females and 57 adult males indicate that the P/C ratio can be used as a measure of metabolism in euthyroid and hyperthyroid states. Statistical analysis of the female series revealed a coefficient of correlation of 0.92. The regression line is: BMR $= 54.7 + 0.22$ P/C. Analysis of the male cases revealed a coefficient of correlation of 0.88, the regression equation being: BMR $= 57.0 + 0.25$ P/C.

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Treatment of Pernicious Anemia with Animal Protein Factor Concentrates of Bacterial Origin

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Animal protein factor concentrates were prepared from the aerobic fermentation of a non-motile gram-negative bacillus isolated from chicken feces. One cc. of one product (A.P.F. 60) had a biologic activity equal to "10 unit liver" when administered to chicks receiving a diet containing 70 per cent soy bean meal. In the *Lactobacillus leichmannii* assay the material had a somewhat lower potency (65% of "10 unit liver"). A second preparation (Normocytin) had a potency of 8 micrograms of B_{12} per cc. by *L. leichmannii* assay and equivalent to 10 micrograms of vitamin B_{12} per cc. by chick assay. Administration of A.P.F. 60 to 5 patients with pernicious anemia in relapse induced a satisfactory reticulocyte response in 4 instances, with an increase of Hb and R.B.C. in all cases. Eight other persons with pernicious anemia in relapse treated with Normocytin showed a satisfactory reticulocytosis and an increase of Hb and R.B.C. Normal levels were observed in 6 cases. One patient from the original group treated with A.P.F. 60 was continued on Normocytin and also reached normal hematologic values. In all instances there was improvement and general well-being of the patients. Megaloblastic bone marrows were converted to normal states. Where neurological changes were present improvement was noted. There was no instance of progression or development of nervous system signs or symptoms. In two patients the Hb and R.B.C. remained below normal levels and are at present under treatment with vitamin B_{12}.

The Significance of RS-T Elevation in Acute Coronary Insufficiency

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The concept of acute coronary insufficiency, as distinct from acute coronary occlusion, has been well established electrocardiographically and pathologically. It represents a disproportion between the demands of the myocardium for oxygen and nutritional elements, and the available supply in the coronary circulation. The mechanism may be inadequate blood flow, increased work of the heart, or impaired oxygenation of the blood. Coronary artery disease is usually present, but the heart may be normal.

There are various degrees of acute coronary insufficiency. Usually the inadequacy of blood or oxygen is transitory and easily reversible, as in simple anginal pain. Anatomical changes in the myocardium are absent or minimal and the process is a functional one. If the coronary insufficiency is severe or persistent, necrosis of the subendocardial layer of the myocardium occurs which may go on to extensive infarction.

In coronary insufficiency the electrocardiogram usually discloses RS-T depression and T-wave inversion, associated with the subendocardial ischemia or necrosis. Accordingly, the electrocardiographic changes may be transitory, or may persist for several weeks.

Coronary artery occlusion usually produces a massive infarct extending to the pericardium. As a result the electrocardiogram presents RS-T elevations and Q waves. The occurrence of such a pattern in coronary insufficiency, while noted occasionally in the literature, has been considered rare. However, in our experience the frequency of RS-T elevation in coronary insufficiency has been great enough to make its differential diagnosis from acute coronary occlusion clinically important.

Our material includes twenty-five cases of acute coronary insufficiency with RS-T elevation instead of RS-T depression. Serial electrocardiographic tracings were obtained, usually including unipolar precordial and extremity leads.

Five of these cases were examined postmortem and no recent coronary occlusion was found. Clinically, all the cases lacked the corroborative evidence of infarction, i.e., elevated white blood count, abnormal sedimentation rate, and fever. Most of the patients had suffered previous coronary occlusion with infarction, usually of the posterior wall. Some, however, had no history or electrocardiographic evidence of previous occlusion with infarction.

In most of our cases the coronary insufficiency with RS-T elevation occurred spontaneously during ordinary activity, or under emotional stress. Other precipitating factors were paroxysmal tachycardia, hemorrhage and shock, and operative procedures including sympathectomy. The RS-T elevation was transitory, disappearing when the precipitating influence was removed. We were able to induce RS-T elevations by exercise and the injection of atropine and adrenalin.

Our data indicate that not infrequently an erroneous electrocardiographic diagnosis of coronary occlusion with infarction may be made in cases of transitory coronary insufficiency if the significance of the RS-T elevation in these cases is not appreciated.

The functional and reversible nature of the RS-T elevations is clearly demonstrated.

Our findings in humans confirm the experimental observations on epicardial and endocardial injury in animals, and contribute further to the understanding of the nature of myocardial injury.

* Dazian Foundation Fellow in Medicine. Sara Welt Fellow in Medicine.

Eugene Hillhouse Pool
1874 - 1949

Eugene Hillhouse Pool — 1874-1949

Dr. Eugene Hillhouse Pool, thirty-eighth President of The New York Academy of Medicine, died on the 9th day of April, 1949, in his seventy-fifth year. Thus ended one of the most distinguished careers in the history of American surgery.

Born June 3, 1874, he was graduated from St. Paul's School, Concord, New Hampshire and entered Harvard in the fall of 1891, receiving his A.B. in 1895. Dr. Pool was graduated in medicine from the College of Physicians and Surgeons, Columbia University, in 1899.

Following a surgical internship at the New York Hospital, 1900-1901, Dr. Pool planned a career in internal medicine. This plan was dramatically altered by an invitation from Dr. William T. Bull to become his associate in an extensive surgical practice. Dr. Bull was one of the master surgeons of his time and probably the first man to confine his work exclusively to surgery. Then followed seven years of constant association with Dr. Bull, both in practice and in surgical writing.

Dr. Pool was an Assistant Demonstrator in Anatomy at the College of Physicians and Surgeons from 1901-1904. He was Attending Surgeon to the French Hospital from 1901-1915 and Consulting Surgeon up to the time of his death. Following an instructorship in surgery at the College of Physicians and Surgeons he became an Associate in Surgery from 1912-1915 and Professor of Clinical Surgery from 1915-1938.

In 1907 Dr. Pool became Associate Attending Surgeon to the New York Hospital and eight years later became Attending Surgeon, which position he held until 1932 when the New York Hospital moved uptown to join with the Cornell Medical College to form the Cornell-New York Hospital Medical Center. Thereafter Dr. Pool became Clinical Professor of Surgery in the Cornell University Medical School and Senior Attending Surgeon to the New York Hospital.

Thus, for more than thirty-five years Dr. Pool was intimately connected with the New York Hospital, an association which brought great fame to hospital and surgeon.

As distinguished Chief of the Second Surgical Division of the New York Hospital Dr. Pool was indefatigable in directing one of the greatest surgical services in the country and became known throughout the surgical world as a master surgeon and preëminent teacher. Ever interested in the problems of the surgical care of patients and in surgical research, he stimulated and was a constant source of inspiration to many students of the art of surgery who have since gone on to their own individual accomplishments and recognition.

His surgical service became a model of its kind, attracting many prominent visitors and his conferences and surgical clinics will forever be remembered by all who were so fortunate as to be participants.

Dr. Pool was an extraordinarily able man. Endowed with a fine mind, he developed it to the highest degree. He was wise in counsel, discerning in judgment, unfailing in leadership, with a limitless capacity for work. When he started anything he never stopped until the job was completed. He gave of himself without stint. He had vision, was a straight thinker and was able to crystallize his thoughts. With it all he was intensely practical and a wonderful judge of men. He knew how to arouse interest in a problem and how to keep that interest. He had a keen sense of kindly humor. He had a love of people and boundless kindness toward people. That is why his patients believed in him, trusted him implicitly and never forgot him.

Dr. Pool's published articles numbered upwards of seventy. He wrote extensively on diseases of the thyroid and parathyroid, heart and pericardium, spleen and gastrointestinal tract. At an early date he required a medical internship as a prerequisite to his surgical internship. He constantly strove to improve surgical technique and was himself a meticulous, gentle operator with a reverence and deep respect for tissues. He instituted routine conferences with the hospital pathologist and

stressed the great importance of a knowledge of pathology in surgery. He refined the abdominal suction tip which bears his name (1909) and was a pioneer in early postoperative exercises which he developed and wrote about while convalescing from an operation upon himself (1913). In 1927, recognizing the entity of psychosomatic syndromes, he arranged for regular ward rounds on his service by a leading psychiatrist from the Westchester Division of the New York Hospital. These are but a few of his extraordinarily important and trail-blazing contributions to surgery.

Dr. Pool's service in World War I was a distinguished one. Going to France with Base Hospital No. 9 (New York Hospital Unit) he became in turn, Chief Surgeon of Evacuation Hospital No. 1, Consulting Surgeon to the 5th Army Corps and finally, Consulting Surgeon to the 1st Army. He was discharged January 30, 1919 with the rank of Lieutenant Colonel and was awarded the Legion d'Honneur, the Distinguished Service Medal and a citation for "meritorious services" from the C.I.C., John J. Pershing.

This distinguished service in World War I was strikingly recalled in World War II when many of his observations, published after World War I—

"The Early Treatment of Gunshot Wounds"—Oxford Surgery, 1921 P745-775.

"Military Surgery of Joints"—Keen's Surgery, 1921 VII, 557.

"Surgery of the Soft Parts and Joints"—S.G.O., 1918 XXVII, 289.

"War Wounds, Primary and Secondary Suture"—J.A.M.A., 1919 LXXIII, 383.

"Treatment of Recent Wounds of the Knee Joint"—Annals of Surgery, 1919 LXX, 266

were of timely interest and of invaluable assistance to the recent World War II surgeon.

Dr. Pool was associated with many hospitals in a consultant capacity, among these being—

Home for Incurables
Central Islip State Asylum
New York Orthopedic Hospital
United Hospital at Portchester
French Hospital
Hospital for Special Surgery (R & C)—where he was Surgeon in Chief, 1934-1936; Surgeon in Chief Emeritus, 1936-1949
New York Infirmary for Women and Children
Monmouth Memorial Hospital
Elizabeth A. Horton Memorial Hospital
North County Community Hospital
Presbyterian Hospital
New York Eye and Ear Infirmary
Harlem Hospital
St. Vincent's Hospital (N. Y. C.)

Despite the press of his exacting hospital duties and an increasing consulting practice, Dr. Pool was active in many professional societies and was accorded their highest honors:

New York Academy of Medicine:—Fellow, 1904; Trustee, 1925-1934 and 1937-1941; President, 1935-1936.

New York Surgical Society—President, 1923-1925.

American Surgical Association—President, 1936.

International Surgical Society.

Southern Surgical Association.

American College of Surgeons—President, 1936.

Society of Clinical Surgery—President 1927-1929.

Detroit Academy of Medicine, Honorary Member.

In 1935 Governor Lehman appointed Dr. Pool Chairman of the Committee to rewrite the medical provisions of the Workmen's Compensation Act. This Act was in answer to the widespread medical abuses revealed by a former committee appointed by Governor Roosevelt. This Committee was an extremely important one and completely rewrote all the provisions of the Workmen's Compensation Act. The governor made the Committee's report his bill and it was passed without any amendments by the State Legislature. Since the passage of the bill, as drawn by Dr. Pool's Committee, it has been changed in only minor particulars and has served as a model for other states which have found it necessary to improve the medical provisions of their workmen's compensation laws.

Dr. S. S. Goldwater, New York City Commissioner of Hospitals, appointed Dr. Pool Administrative Consultant in Surgery for the City hospitals in 1934.

Dr. Pool was a life trustee of Columbia University, an Honorary Governor of the New York Hospital, a life Trustee of the Children's Aid Society and a former Trustee of St. Paul's School, Concord, New Hampshire. In 1944 he was awarded Columbia University Alumni Federation's Gold Medal for distinguished contributions to the University.

Dr. Pool loved sports and was an accomplished skater, above average golfer and played excellent tennis. For many years he looked forward to annual fishing and shooting trips. When he had the opportunity in his busy life he played as he had worked, with great skill, enthusiasm and with his characteristic will to win.

Dr. Pool's clubs were the Links (N. Y.), Piping Rock (L. I.), and the Harvard Club of which he was a former President. His life was crowded with activity and accomplishments.

Outside of his profession Dr. Pool's outstanding interest was that of a collector. Being a great, great grandnephew of Captain James Lawrence of "don't give up the ship" fame, he brought together over a period of years a remarkable collection of memorabilia and other objects concerning Captain Lawrence. Today this collection may be seen in one of the galleries of the New York Historical Society.

It is rare for a doctor to receive and deserve an editorial in a leading New York paper. The Evening Sun, on April 13, 1949, paid Dr. Pool this superb tribute:

"It would be impossible to estimate the number of persons who owe their lives to Dr. Eugene Hillhouse Pool. Even a complete roster of all who came under his hands as patients would only begin to tell the story. To those would have to be added the patients of thousands of men, surgeons today in all parts of the world, who gained their skill from watching the amazing hands of this master surgeon and from listening to his sound advice on how best to repair broken bodies. Among a whole generation of surgeons there is no prouder boast than that of having been a pupil of Eugene Pool, that extremely strict taskmaster who demanded rigid perfection in his students but who spared nothing in seeking to attain it himself. If his capabilities in the operating room alone were taken into account, the world would be heavily in his debt. Coupled with them, however, were an unswerving integrity, a tremendous interest in all matters pertaining to the medical profession and the fine instincts that mark the true gentleman whatever his field of endeavor. As President of the Academy of Medicine in a period when it was seeking to shape its future course, he did much to set it on the road to its present high estate. A man who habitually shunned the spotlight, Dr. Pool on his death left little material for biographers. More important, he left a shining example for doctors, present and future."

Dr. Pool was divorced from his first wife, Mrs. Esther Hoppin Pool, in 1930 and his second wife, Mrs. Kitty Lanier Harriman Pool, died in 1936. Surviving are his third wife, Mrs. Frances Saltonstall Pool, two sons by his first marriage, Dr. J. Lawrence Pool and Beekman H. Pool, a brother, Harry Henry Pool and a sister, Mrs. Sophie Duer.

In his death on April 9, 1949 the profession lost a great leader, the community a great force, his associates a great friend.

"The soil out of which such men as he are made is good to be born on, good to live on, good to die for and good to be buried in."

FRANK J. McGOWAN

TWENTY-SECOND GRADUATE FORTNIGHT

of

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October 10 to 21, 1949

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ADVANCES IN DIAGNOSTIC METHODS

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Afternoon Hospital Clinics

Evening Addresses and Motion Picture Presentations

Scientific Exhibits

Registration cards will be furnished to all Fellows of the Academy without application. Registration cards will be sent to other applicants upon receipt of check for ten dollars, payable to The New York Academy of Medicine. Address, Secretary, Fortnight Committee, The New York Academy of Medicine, 2 East 103 Street, New York 29, N. Y.